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CONTENT

EDITORIAL

Following a hectic few months, here is the next (slightly delayed) issue of the Journal. Once again a very large thank you to those reviewers that have assisted me over the last six months; your valuable time and positive comments are much appreciated by me and our authors (whether published or declined). Giving constructive feedback to those who have worked for long hours on manuscripts helps to maintain standards and raise motivation. It has also been extremely pleasant to chat online with various reviewers across the world. The new website is also up and running thanks to Herman and Zephne Bernitz and colleagues.

Over the last few months mass fatality incidents have claimed our attention and caused chaos and destruction around the world. Over here, in the southern hemisphere, we have had our share of incidents to include; flooding in Queensland, the Pike River mining explosion and the series of earthquakes that destroyed much of Christchurch with the loss of 182 lives. The aftershocks continue to distress the residents and cause further damage to buildings, land and livelihoods. Whilst deployed with the New Zealand dental DVI team, we received the shocking news of the earthquake and tsunami in Japan. The disaster response and identification teams can and do make a difference to these incidents and being prepared is essential. Errors in identification cause additional distress to relatives and friends of the deceased and accuracy and quality assurance is essential. Turbulent times need effective teams.

The recent New Zealand Society of Forensic Odontology meeting in early May focused on the Christchurch disaster response and the subsequent dental debrief was most worthwhile to discuss lessons learned, standard operating procedure updates and future responses. The friendships gained from working within the disaster teams are always rewarding; a particular bond is felt by all with the hope that we have assisted in some small way.

The 19th IAFS meeting is to be hosted in Funchal, Madeira (12-17 September). This promises to be a most interesting meeting with a dedicated forensic odontology section and workshop. Is there a better place to learn, exchange ideas, update your continuing professional development, renew old friendships and make new friends? Book a few extra days to enjoy the history, levada walks and fortified Portuguese wine. Unfortunately, I may have to miss this opportunity and stay on this side of the world for the Australian Society of Forensic Odontology Symposium in Darwin in October. We are fortunate that forensic odontology has such amazing international conference opportunities and members prepared to put in the effort to ensure that the meetings are organised and information is shared.

The Journal is receiving a steady influx of manuscripts to be considered for publication, however, some member countries are not contributing yet! I am sure that someone in each country has a fascinating case that they would like to share with us? Our December issue is not too far away – please start work immediately! We are now finally back with PubMed/ Medline which has taken a surprising amount of work this year; thank you to Charles Maybury, our assistant editor, for all his technical skills with this matter.

I look forward to hearing from you and send kind regards for the summer or winter wherever you are.

Judy Hinchliffe (BDS, Dip F Od, Hon FFFLM)
Editor

AGE ESTIMATION IN AN INDIAN POPULATION USING PULP/TOOTH VOLUME RATIO OF MANDIBULAR CANINES OBTAINED FROM CONE BEAM COMPUTED TOMOGRAPHY

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ABSTRACT

The present study assessed the suitability of pulp/tooth volume ratio of mandibular canines for age prediction in an Indian population. Volumetric reconstruction of scanned images of mandibular canines from 140 individuals (aged ten - 70 years), using computed tomography was used to measure pulp and tooth volumes. Age calculated using a formula reported earlier for a Belgian sample, resulted in errors > ten years in almost 86% of the study population. The regression equation obtained for the Indian population: Age = 57.18 + (- 413.41 x pulp/tooth volume ratio), was applied to an independent control group (n = 48), and this resulted in mean absolute errors of 8.54 years which was significantly ($p < 0.05$) lower than those derived with the Belgian formula. The pulp/tooth volume ratio is a useful indicator of age, although correlations may vary in different populations and hence, specific formulae should be applied for the estimates.

(J Forensic Odontostomatol 2011;29:1:1-6)

Keywords: Forensic science, forensic odontology, age estimation, Indian, pulp volume, tooth volume, cone beam computed tomography

Running ttle: Age estimation using pulp/tooth volume ratio and cone beam computed tomography

INTRODUCTION

The identification of age in anthropology and forensic medicine is sometimes difficult, but nonetheless important. Several methods based on the analysis of teeth have been reported in the literature to estimate the unknown age of individuals. The most widely used methods include the analyses of Gustafson[1] and Johanson,[2] as well as the assessment of dentinal translucency[3] and of cementum annulations.[4,5] Some methods are destructive and are therefore not appropriate for living individuals. In anthropology,[6] the analysis of dental wear[7] is the most commonly used method.

Examination of the pulp space offers new opportunities in dental age identification. A commonly used method found to be effective is the evaluation of secondary dentine apposition.[8] This apposition is a continuous, age-associated process, which alters the size of the pulp chamber and is only modified under pathological conditions (for example, caries). Various methods have been developed to study the size of the pulp chamber, including tooth cross-sections and radiographs. Both panoramic[8] and periapical radiographs[9] have been used to assess the pulp/tooth area ratio of maxillary canines. The primary disadvantage of radiographs is that they are two-dimensional projections which are subject to considerable magnification and distortional errors. Therefore, a simultaneous assessment of the mesio-distal and bucco-lingual dimensions of teeth has been recommended. Computed tomography (CT) is the ideal and most accurate method to evaluate the pulp/tooth volume ratio[10] and a previous study reported on dental age estimation based on microfocus CT images of extracted teeth.[11]

Internal complexities of the root canal seem to be genetically determined and are considered important in anthropology[12] and the same can be expected to apply to the relative proportion of the pulp and tooth volumes. However, there are very few reports on the use of CT to determine the pulp/tooth volume ratio of different

populations so that a valuable data set is missing for forensic medicine. An extensive review of literature showed that there was only one report on the use of Cone Beam Computed Tomography (CBCT) to determine the pulp/tooth volume and correlate it to age.[10] Therefore, the aim of the present study was to analyze the pulp/tooth volume ratio of canines in an Indian population by CBCT and correlate it to age. The null hypothesis was that there is no such correlation.

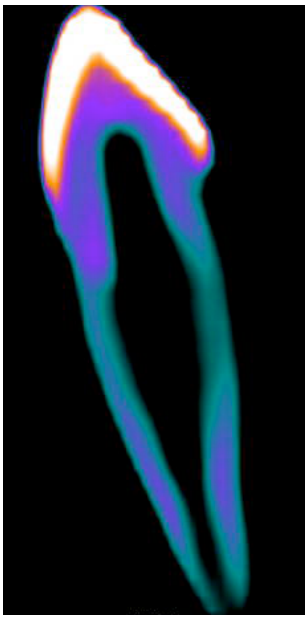

Fig. 1: *Reconstructed CT image of tooth with tooth volume marked.*

MATERIALS AND METHODS

One hundred and eighty-eight intact, non-carious mandibular canines were collected from dental practitioners across the Indian subcontinent. The age of the subjects ranged between ten and 70 years and was evenly distributed across different age groups (Table 1). Subjects were divided into a study sample of 140 individuals and a control group of 48, taking care that age-distribution was similar in both groups. All teeth were obtained from indigenous Indians and no specimens from other ethnic minorities were included. The process of collection was performed by a team of practitioners (who were informed about the aims of the study) and every tooth was accompanied by a case record stating the ethnicity and age of the patients. Carious teeth, teeth with restorations and excessive wear were excluded. The teeth were washed under tap water immediately after extraction and stored in distilled water with thymol iodide crystals (Titan Pharma, Mumbai, India) until the collection was completed after about two months. Then the specimens were washed thoroughly under tap water and immersed in 2.5 % sodium hypochlorite (Prime Dental Products, Mumbai, India) for 30 minutes to remove adherent soft tissue.

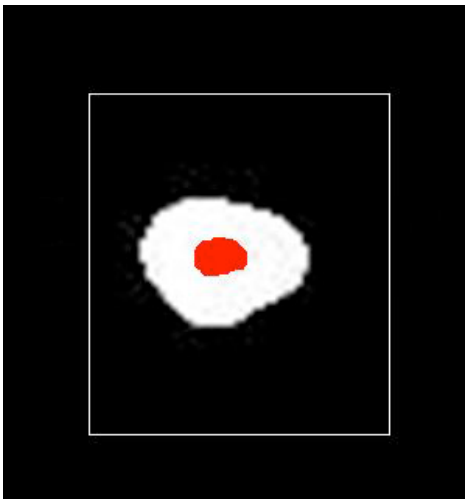

Fig. 2: *A cross-sectional slice of the tooth obtained by CT, outlining the pulp space.*

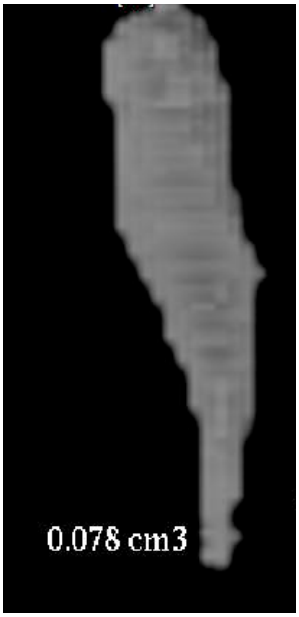

Fig. 3: *Volumetric reconstruction of pulp space.*

All the teeth were imaged by a Cone Beam Computed Tomography scanner (3DAccuitomo, J Morita Corporation, Osaka, Japan) with a constant slice thickness of 250 µm and exposure conditions of 140 kV and 550 mA. The scans were evaluated using both cross-sections and longitudinal sections. Volume rendering and multiplanar volume reconstruction (Fig. 1) was performed with the Advantage Windows workstation (GE Systems, USA). Using a method described previously[10] the pulp/tooth volume ratio was determined by two independent examiners who were blinded with regard to personal data of the patients (Fig. 2-4). Age was then calculated using the linear regression equation derived by Yang et al. for mandibular canines.[10]

Fig. 4: *Superimposition of pulp volume reconstruction on tooth volume image.*

Table 1: *Sample distribution across age-groups and sexes.*

Age group (years)	Males	Females	Total
Study group			
10-20	04	03	07
21-30	11	11	22
31-40	15	13	28
41-50	14	13	27
51-60	14	13	27
>60	15	14	29
Total	**73**	**67**	**140**
Control group			
10-20	4	4	8
21-30	4	4	8
31-40	4	4	8
41-50	4	4	8
51-60	4	4	8
>60	4	4	8
Total	24	24	48
Grand total	**97**	**91**	**188**

All the results from the measurements along with additional information such as: type of tooth (right or left canines) and the individual's age and gender, were entered in a spreadsheet (Microsoft Excel 2007). Inter-rater agreement (reproducibility) was determined using the data of the two examiners. Intra-rater agreement (repeatability) was assessed by having both the examiners evaluate one half of the CBCT images at two separate sessions. Results from

repeated measurements were subjected to a paired t-test to assess potential systematic intra- and inter-observer errors. The intraclass correlation coefficient was calculated to estimate the reliability of the measurements recorded by the two examiners. In addition, the size of the measurement errors was calculated as $\sqrt{(SUM(d^2)/2N)}$, where d are the differences between double measurements and N the number of double measurements. From the differences between the actual and predicted ages the mean absolute error (MAE) was calculated to obtain a measure of the accuracy of the age prediction.[13] For the same purpose, the percentage of estimates with deviations of more than ± ten years, which is considered as ''acceptable'' in forensic age prediction,[14] was also determined.

As this analysis yielded unexpectedly large errors, a linear regression analysis was performed with the pulp/tooth volume ratios and ages obtained from the Indian sample (SPSS 10.0, SPSS Inc., Chicago, IL, USA). The regression equation so derived was then applied to the control sample (n = 48) to estimate ages and determine the errors of the estimates as described above. The two data sets obtained from the study and control group finally allowed comparisons of the accuracy of the age predictions with the Indian and Belgian formulae.[10]

RESULTS

Differences between repeated measurements at two time points and of different examiners were not statistically significant ($p > 0.05$). There were no significant differences between the samples in terms of type of tooth (right or left canines) or gender of patients. The value of the intraclass correlation coefficient was very high ($\rho=0.99$). Measurement errors produced with analyses in two separate sessions and of the two examiners were 0.75 and 1.23 respectively. The use of the formula established for the Belgian population[10] in the study group produced an MAE of 15.34 years in 72.81% of the cases. Age estimates were within ±10 years of actual age in 27.09% of the cases.

Regression analysis yielded a statistically significant but moderate negative correlation between pulp/tooth volume ratio and age (Pearson's correlation coefficient $r = -0.63$; $p < 0.05$; Fig 5) and the following linear regression equation: Age = 57.18 + (- 413.41 x PTV), where PTV is the pulp/tooth volume ratio. Application of this formula to the control group yielded an MAE of 8.54 years with 29/40 (72.5%) estimates lying within ± ten years of actual age. In contrast, the use of Yang's formula[10] in the control group produced an MAE of 14.78 years, which was significantly different ($p < 0.05$) from the MAE obtained with our new formula.

DISCUSSION

This study indicates that the pulp/tooth volume ratio of human mandibular canines is useful for age predictions, but that respective formulae vary between different populations and have to be adapted: in this case for the Indian population. The use of morphological characteristics of the teeth is considered to yield more reliable age estimates than most other methods.[15] The assessment of pulp/tooth area ratio and pulp/tooth volume ratio are methods to indirectly quantify secondary dentine deposition. Secondary dentine deposition is an age-associated process along internal tooth surfaces which can be considered well protected against environmental influences. Consequently, evaluation of this parameter may have the potential to eliminate at least part of the effects of external factors.

An earlier report on the application of pulp/tooth area (rather than volume) ratio to estimate age concluded that the formula which had been derived for an Italian population could be applied to Indians as well.[16] However, the results of our study do not support this conclusion, but rather suggest that a formula devised for one population may not be applicable for another. This may be due to anthropological differences between various ethnic populations, but could also be attributed to the fact that pulp/tooth area ratios are calculated from radiographs, which are two-dimensional representations of a three-dimensional object.

Cone Beam Computed Tomography (CBCT) was used to calculate the pulp/tooth volume ratio in our study, for two reasons: (1) The analysis of the volumes of the pulp chamber and tooth is more reliable than calculation of area ratios, possibly because secondary dentine formation may not be uniform along all pulp surfaces and, hence, measurements of

projected areas could give an incorrect impression of the extent of this process; (2) CBCT is an accurate technique for studying the anatomy of the pulp chamber and root canal system[17] which provides three-dimensional volumetric information of the teeth of living individuals by a single scan and can be operated non-destructively. A similar method has been used for age identification of a Belgian population.[10] The results of this present study show that the formula, though accurate for the Belgian population, did not hold true for the Indian subjects, yielding errors of age estimates exceeding ten years, which is considered unacceptable in forensic age identification.[18] The Indian population is generally considered to be a hybrid of several ethnic groups with characteristics of Caucasian, Mongoloid and Negroid races, which is generally referred to as the Dravidian group.[19] If anthropological differences also existed with respect to the relationship between the pulp/tooth volume ratio and age, these could account for the different outcomes of age predictions in the Belgian and Indian population and further justify the use of population-specific equations.[20] In fact, the correlation derived from the data of our Indian sample was much higher than that demonstrated in the previous report.[10]

CONCLUSION

The present work showed that with a modification of Yang's formula acceptable age predictions are possible for the Indian population using pulp/tooth volume ratio measurements (of mandibular canines) from Cone Beam Computed Tomography. However, caution should be exercised when extrapolating these results to practical applications. Further research including different types of teeth, thus facilitating the development of multiple regression models could possibly enhance age prediction; additionally further studies should also analyze the possible impact of wear and tertiary dentine deposition on the pulp/tooth volume ratio.

ACKNOWLEDGEMENTS

The authors wish to thank Dr. Robert B. J. Dorion, Director of Forensic Dentistry, Laboratoire de Sciences Judiciaires et de Médecine Légale, Ministry of Public Security for the Province of Quebec, Montreal, Quebec, Canada; Dr. Harry H. Mincer, Professor and Director of Oral and Maxillofacial Diagnostic Services, University of Tennessee College of Dentistry, for pre-reviewing the manuscript and for their scientific advice, and Dr. Matthias Zehnder, Universität Zürich, for his valuable guidance in the statistical analysis.

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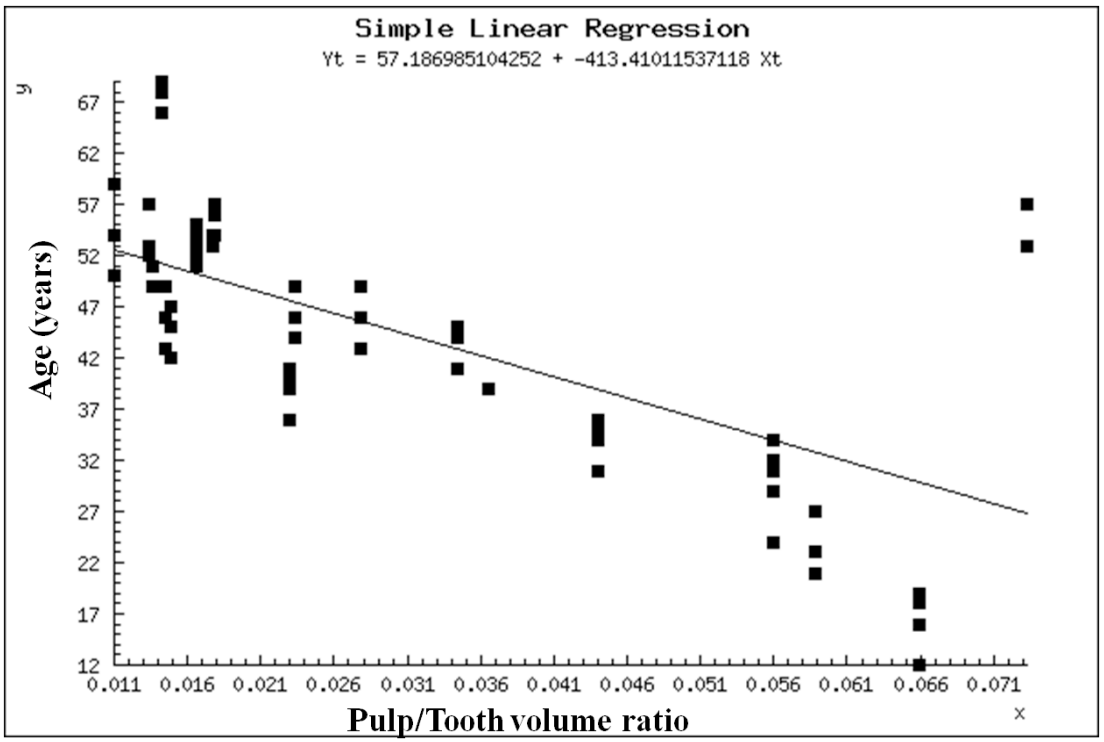

Fig. 5: *Graphical representation of age as a function of the pulp/tooth volume ratio.*

STATURE ESTIMATION BY CARREA'S INDEX AND ITS RELIABILITY IN DIFFERENT TYPES OF DENTAL ALIGNMENT

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ABSTRACT

Stature is a measurable feature of the body, useful in human identification, which may include or exclude an individual from a missing persons list. The aim of this study is to analyze the Carrea´s index for stature estimation in dental arches with normal dentition, crowding and diastema. Plaster casts of 51 students of the Federal University of Paraíba were analyzed. Each hemiarch was divided according to the dental position, and the elements were measured with divider and digital calipers. Considering the normal and crowded dentition, the Carrea´s index presented a satisfactory success percentage, between 72.2% and 95.2%, with no statistically significant difference between sexes or between right and left sides. The presence of diastema reduced the number of matches to less than 62.5%. It was concluded that the Carrea´s index is a reliable method for height estimation in arches with normal and crowded dentitions, useful in males and females, and in the right and left sides. However, the method was not efficient in hemiarches with diastema.

(J Forensic Odontostomatol 2011;29:1:7-13)

Keywords: Forensic Anthropology, anthropometry, forensic dentistry, dental arch, stature measurement

Running title: Stature estimation, Carrea´s index, reliability in different types of dental alignment

INTRODUCTION

Anthropometry is the branch of physical anthropology that studies the quantitative variations of human features, such as stature.[1-5] In a forensic context, among the information potentially collected from human remains, estimated stature can be an important feature to be added to the criteria, helping to narrow the search from missing person's data.[6-8]

Stature is the total height of a person, and it varies according to sex, age, ancestry, individual development, and hormonal influence. In the deceased 9 to 17mm must be added to the measurement of the body in the supine position due to the natural flattening of the intervertebral discs, varying according to sex, ancestral background, nutrition, body composition, climate, and day length.[9,10] In a full skeleton, the heights of the body segments that contribute to stature are summed, and added to correction factors according to sex and calculated height.[11] The stature, however, is often estimated from various parts of the body, and more commonly from long bones.[1,6,8,12-15] Nevertheless, in many investigations of human remains, not all the bones are present; possibly nothing but the skull and mandible may be recovered. In that regard, the examination of skull and teeth becomes very important, and the stature of the subject can still be estimated based on the proportionality with tooth dimensions. Carrea created a formula that allows the stature to be estimated from measurements of the lower anterior teeth.[2,16-18]

The aim of this paper is to validate Carrea´s index, testing the method reliability in different types of tooth alignment, considering the increasing prevalence of malocclusion among populations.[19]

MATERIALS AND METHOD

Sampling

For this research, data were taken from 51 undergraduate students from the Federal University of Paraíba: 24 males and 27 females, aged between 18 and 30 years. Casts of the lower dental arch of each subject were obtained from alginate impressions immediately poured in plaster. The stature was measured with an anthropometer, by making the subject stand erect on the horizontal plane, barefooted, in the anatomical position according to the Frankfurt plane, in inspiratory apnoea, aligning the posterior surface of heels, pelvic girdle, scapular girdle, and occipital region to the vertical plane. The stature was measured with the rod of the anthropometer

in contact with the vertex. All measurements were performed by a single investigator, in the morning period.

Data collection

For examining the casts, each hemiarch was considered separately, equaling 102 inferior hemiarches, which were divided according to the dental alignment into three groups: normal (n=41), crowded (n=43), and diastema (n=18). The measurements of each hemiarch were performed as described by Carrea (1939):

Arch: the sum of the mesiodistal diameters of the inferior central incisor, lateral incisor, and canine, measured by the labial surface (Figure 1).

Chord: the linear distance between the ends of the arch, represented by the mesial edge of the central incisor and the distal edge of the canine on the same side, measured by the lingual surface (Figure 1).

The intra-examiner test was performed in the measurement of the arch and chord of twelve casts. As ascertained by Carrea, the measurements of the arch and the chord, can be used to estimate the individual stature by the so named Carrea´s index, calculated by the formula below:

$$\text{Maximum stature} = \frac{\textbf{\textit{arch}}\ (\text{in mm}) \times 6 \times 3{,}1416 \times 100}{2}$$

$$\text{Minimum stature} = \frac{\textbf{\textit{chord}}\ (\text{in mm}) \times 6 \times 3{,}1416 \times 100}{2}$$

The mesiodistal diameter of each dental element was taken by the use of a divider caliper (ICE, São Paulo, Brazil) and a millimeter scale (Angelus, Paraná, Brazil). The chord was measured with a digital caliper (Digimess, São Paulo, Brazil). The estimation obtained with the formula was then compared to the real stature of each individual. To evaluate the intra-examiner reliability, twelve hemiarches were re-measured after an eight days interval.

Statistical analysis

All data were inserted in an Excel file, and analyzed using Statistical Package for the Social Sciences (SPSS) version 13.0. The intra-examiner reliability was tested by the Kappa index, and the remaining data, by the Pearson´s Chi-Square test, and Fischer´s exact test, at a 95% confidence interval.

RESULTS

Regarding the intra-examiner test, there was a reliable result by the Kappa index (0.74), with a confidence interval of 95%. The descriptive statistics relating to each kind of dentition among the sexes is shown in Table 1. In addition, the frequency of each type of dental alignment related to the bilateral hemiarches, considering the 51 inferior casts, is shown in Figure 2.

The analysis of Carrea´s index applied to the hemiarches with normal dental position has shown a higher percentage of success in males (81.3%) than in females (76.0%), although without statistically significant difference (p=1.00), as shown on Table 2. The same thing occurred when left and right sides were compared in each dental arch (Table 3). Though the success rates were higher for right side (82.6%) comparing to the left (72.2%), there was no statistical difference between them (p=0.471).

Considering the group of crowded teeth, Tables 4 and 5 demonstrate a higher rate of success for females (95.2%) and for the right side (90.0%), against the results for males (77.3%) and for the left side (82.6%). However, this difference in percentage has not proven to be statistically significant (p=0.185 and p=0.669 for sex and side, respectively).

As for the group with diastema, there was a balanced distribution of success (50.0%) for both sexes (p=1.00), as can be seen on Table 6. By examining both sides (Table 7), the success rates for the right side (62.5%) were higher than the left (40.0%), although the difference was not statistically significant (p=0.637).

DISCUSSION

The Carrea´s index for normal arches, as originally described,[18] has shown significant rates of success in both sexes with no statistical difference between them. The same result was seen on left and right hemiarches, demonstrating that the method can be applied on both sides, without affecting the outcome.

It is worth noting that the studies published by Carrea date back to 1920 and 1939[17,18] when methodological patterns were not observed by the authors, and papers often lack important information. Therefore, there is no Carrea´s paper describing how exactly he got to his formula. It is known, however, that he analyzed dry skulls and mandibles, instead of living people, and does not mention any kind of alignment disorder as a possible cause of failure of the method. His theories discuss the measurements of dental elements, the concept of arch and chord, and its relation with measures of mandible, face and skull. The present study upgrades the concepts stated by Carrea, as it put to the test his stature estimation method in living people of a contemporary population, in which it has shown to be reliable. The variation of the final outcome in dental arches with crowding and diastema was also assessed for the first time in this research.

The sample analyzed was conventionally composed of students of the Federal University of Paraíba, which, as a federal educational institution, is maintained by the State, and not charged to the students. Therefore, studying in a federal educational institution in Brazil does not mean economical prosperity. As to the ancestry, Brazil is known to host one of the most mixed populations in the world. After 510 years of interethnic crosses between Amerindians, Europeans and Africans, Brazilian people should be assessed as one of a kind. Genetic details can be seen in the studies of Pena et al,[20] Santos et al[21] and many others.

In contradiction of Croce & Croce Júnior,[22] who discourage the use of Carrea´s index for crowded arches, this study has obtained significant success rate on this kind of dentition, for both males and females, and both sides, sometimes even higher than in cases with a normal dentition.

Cavalcanti et al[23] had used two methods of measuring cast dental elements for estimating height through the Carrea´s index. In the one he called conventional, the arch was measured with a millimeter tape, and the chord, with a caliper; in the modified method, the arch and the chord were measured with a divider caliper. The study observed that, in the modified method, the rate of success was higher in males (100%) than in females (93.3%), with equivalent rates in both sides. The conventional method has shown lower rates of matching: 35 and 45% for males (right and left sides), and 36.7 and 50% for females (right and left sides, respectively). It should be noted that normal and crowed arches were analyzed together, with no distinguishing between them. The paper does not provide the result of statistical tests between sexes.

There was a consistently higher correlation between stature estimation and the right hemiarch in this study, even though not statistically significant. This finding is in contradiction to the results of Cavalcanti et al[23] which has shown equivalent success rates for both sides by the modified method, and higher correlation to the left hemiarch by the conventional method. As stated by Carrea,[24] any hemiarch can be used to estimate the stature, considering the principle of bilateral symmetry, accepting small variations as normal asymmetries. The statistically insignificance of the variation found by this research sustains Carrea´s theory.

In the study developed by Silva,[16] the chord was measured by a caliper, and the arch by a millimeter tape. The author reported that the real stature of the sample matched the estimations in 70% of the cases. However, there was no distinguishing between normal arches, crowding or any other kind of anomaly. Both hemiarches and sexes were examined together, with no distinction.

Regarding the hemiarches with diastema, the low success rate was caused by the increasing of the chord value, affecting the final result. In these cases, the chord, that predicts the minimal height, was higher than the arch, which estimates the maximum height. The occurrence of these events, in which the error was in the minimal stature, may have been the reason for the low success percentage.

Given the methodology used and results obtained from the sample analyzed, it appears that the Carrea´s index can be used both for males and females as well as in right and left sides, being reliable in the arches with normal and crowded dental positioning. For the hemiarches presenting diastemas, the method was shown not be accurate due to the low rates of success found.

CONCLUSION

Height is a usefel element in human identification and its estimation is not an easy task. The Carrea´s index for stature estimation is a convenient, simple and inexpensive method, and can provide valuable information to the forensic investigation when dental remains are present.

Since the index considers the metric relation between mesiodistal width of anterior elements, the presence of diastema affects the final result, making the estimated minimum stature higher than the maximum stature. Therefore, in hemiarches with diastema, the method presented the lower rate of success.

It should be noted that any human identification method must be tested and validated on local samples. The current level of human variation, especially on interethnic admixed groups, challenges the researchers to keep updated information about local populations.

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TABLES:

***Table 1*:** *Descriptive statistics relating to sex, kind of dental alignment and side of hemiarch.*

Alignment / Sex		Normal		Crowded		Diastema	
		Right	Left	Right	Left	Right	Left
Male		10	06	10	12	04	06
	Total	**16**		**22**		**10**	
Female		13	12	10	11	04	04
	Total	**25**		**21**		**08**	

Table 2: *Distribution of correct and incorrect estimations for arches with normal dentition, according to sex*.

Sex	Correct / Incorrect						*p* value[1]
	Correct		Incorrect		Total		
	n	%	n	%	n	%	
Male	13	81.3	3	18.8	16	100.0	1.000
Female	19	76.0	6	24.0	25	100.0	
TOTAL	**32**	**78.0**	**9**	**22.0**	**41**	**100.0**	

(1): According to Fischer´s exact test.

***Table 3*:** *Distribution of correct and incorrect estimations for arches with normal dentition, according to side.*

Side	Correct / Incorrect						*p* value[1]
	Correct		Incorrect		Total		
	n	%	n	%	n	%	
Right	19	82.6	4	17.4	23	100.0	0.471
Left	13	72.2	5	27.8	18	100.0	
TOTAL	**32**	**78.0**	**9**	**22.0**	**41**	**100.0**	

(1): According to Fisher´s exact test.

Table 4: *Distribution of correct and incorrect estimations for crowded arches, according to sex.*

Sex	Correct / Incorrect						*p* value[1]
	Correct		Incorrect		Total		
	n	%	n	%	n	%	
Male	17	77.3	5	22.7	22	100.0	0.185
Female	20	95.2	1	4.8	21	100.0	
TOTAL	**37**	**86.0**	**6**	**14.0**	**43**	**100.0**	

(1): According to Fisher´s exact test.

Table 5: *Distribution of correct and incorrect estimations for crowded arches, according to side.*

Side	Correct / Incorrect						*p* value[1]
	Correct		Incorrect		Total		
	n	%	n	%	n	%	
Right	18	90.0	2	10.0	20	100.0	0.669
Left	19	82.6	4	17.4	23	100.0	
TOTAL	**37**	**86.0**	**6**	**14.0**	**43**	**100.0**	

(1): According to Fisher´s exact test.

Table 6: *Distribution of correct and incorrect estimations for arches with diastema, according to sex.*

Sex	Correct / Incorrect						*p* value[1]
	Correct		Incorrect		Total		
	n	%	n	%	n	%	
Male	5	50.0	5	50.0	10	100.0	1.000
Female	4	50.0	4	50.0	8	100.0	
TOTAL	**9**	**50.0**	**9**	**50.0**	**18**	**100.0**	

(1): According to Fisher´s exact test.

Table 7: *Distribution of correct and incorrect estimations for arches with diastema, according to side.*

Side	Correct / Incorrect						*p* value[1]
	Correct		Incorrect		Total		
	n	%	n	%	n	%	
Right	5	62.5	3	37.5	8	100.0	0.637
Left	4	40.0	6	60.0	10	100.0	
TOTAL	**9**	**50.0**	**9**	**50.0**	**18**	**100.0**	

(1): According to Fisher´s exact test.

FIGURES

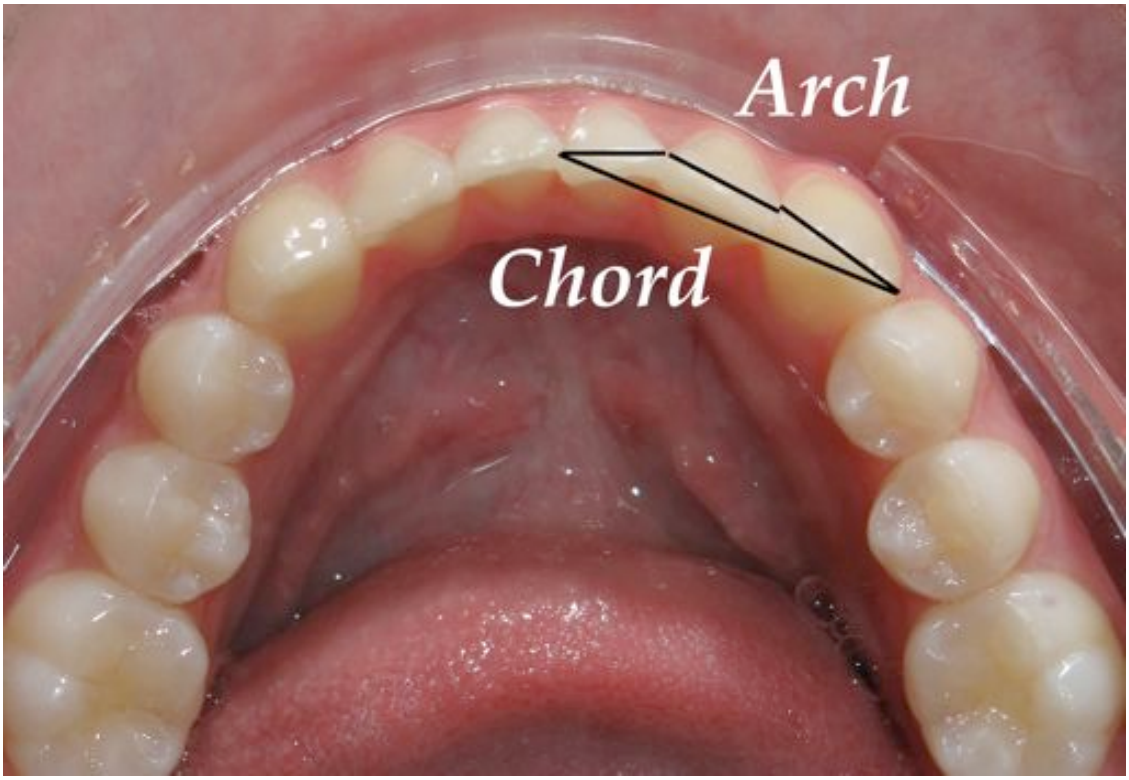

Fig. 1: *Illustrating the measurements of arch and chord.*

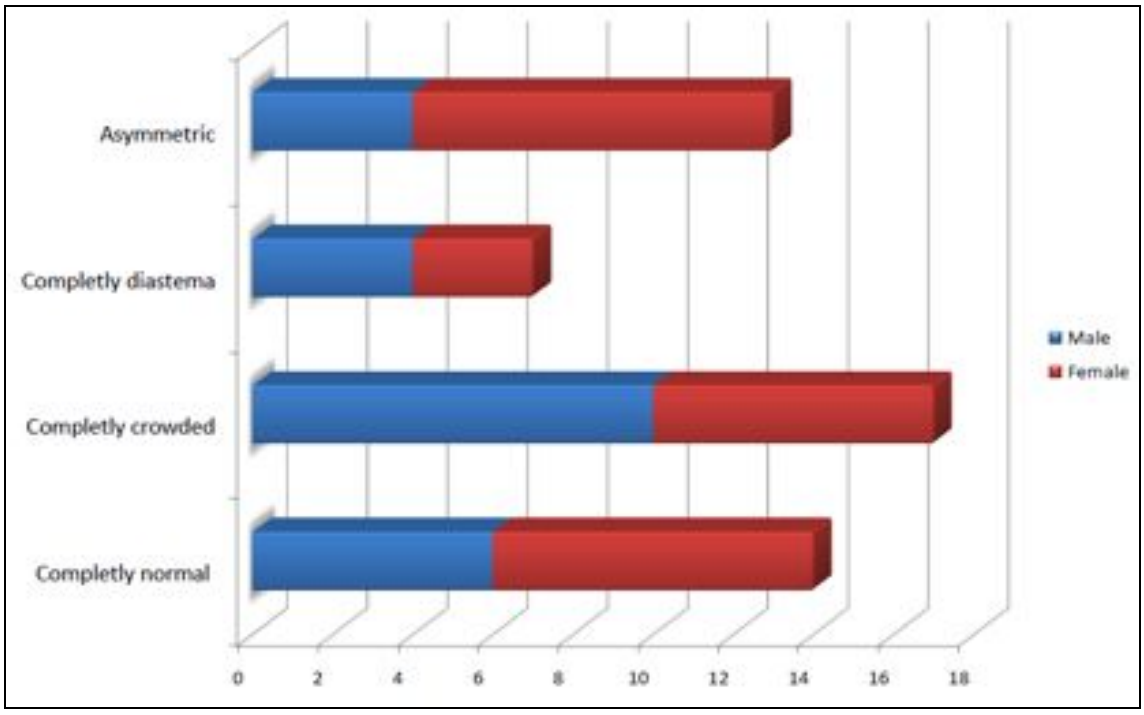

Fig. 2: *Frequency of each type of dental alignment related to the bilateral hemiarches, considering the 51 inferior casts.*

RUGAE PATTERNS AS AN ADJUNCT TO SEX DIFFERENTIATION IN FORENSIC IDENTIFICATION

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ABSTRACT

It is widely acknowledged that in some forensic situations there are limitations to identification of the deceased by fingerprints, DNA and dental records. Palatal rugae pattern of an individual may be considered as a useful adjunct for sex determination for identification purposes. The aim of this study was to identify and compare the rugae pattern in Indian males and females, as an additional method of differentiating the sexes in various postmortem scenarios. Dental stone casts of 120 Indians: 60 males and 60 females were obtained. The method of identification of rugae patterns was that of Thomas and Kotze (1983) and Kapali et al (1997) which includes the number, length, shape and unification of rugae. Our study revealed no significant difference in the total number or various length measurements of rugae between the two sexes which conforms to previous results. However, in terms of the different types of rugae shape, the converging type of rugae were statistically greater in number in females whilst the circular type of rugae were statistically greater in number in males, which contrasts with earlier studies. The use of logistic regression analysis (LRA) enabled highly accurate sex prediction (>99%) when all the rugae shapes were analyzed. It may be concluded that rugae pattern through the use of LRA can be an additional method of differentiation between the Indian male and female and assist with the identification process in conjunction with other methods such as visual, fingerprints and dental characteristics in forensic sciences.

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Keywords: Palatal rugae, rugae pattern, forensic identification, sex assessment, logistic regression analysis

Running title: Sex assessment using palatal rugae patterns

INTRODUCTION

Forensic Odontology is a specialty in dentistry which occupies a primary niche within the total spectrum of methods applied to medico-legal identification. Forensic odontology can be defined as a branch of dentistry which deals with the appropriate handling and examination of dental evidence and with the proper evaluation and presentation of dental findings in the interest of justice.[1]

Identification of the deceased is a prime requisite for certification of death and for personal, social and legal reasons. DNA, fingerprint and dental record comparisons are the most commonly used scientific methods of forensic identification.[2,3] Limitations to the use of fingerprints occur in situations where the hands are charred or mutilated and while teeth are more durable, identification using dental records may also prove to be inconclusive, since many antemortem dental records may be inaccurate or incomplete.[4] Also, additional dental treatment might have been performed in the time interval between the creation of a dental record and death of the individual.[5]

Palatal rugae have been shown to be highly individual and consistent in shape throughout life.[6-10] The anatomical position of the rugae inside the oral cavity (surrounded by cheek, lips, tongue and the buccal pad of fat) also give some protection in cases of trauma or incineration. When identification of an individual by other methods is difficult, palatal rugae may thus be considered as an alternative source of information (usually if comparative material is available) enabling the search field to be narrowed.

Palatal rugae in mammals are transversely running crests, which are exclusively formed by the mucosa of the hard palate except where an ossified base can be distinguished. According to the Glossary of Prosthodontic Terms-8, rugae are anatomical folds or

wrinkles (usually used in the plural sense); the irregular fibrous connective tissue located on the anterior third of the palate.[11] They are also called “plica palatinae” or “rugae palatine.”

It is assumed that the rugae facilitate food transport through the oral cavity, prevent loss of food from the mouth, and participate in food crushing. Because of the presence of tactile and gustatory receptors, rugae contribute to perception of taste, mechanical food qualities, and tongue position.[12]

Despite being protected by their internal position within the head, some events can contribute to changes in rugae pattern, including trauma, extreme finger sucking in infancy and persistent pressure with orthodontic treatment and dentures. In one study, it has been reported that no two palates are alike in their configuration and that the palatal print did not change with time or age. Even between twins, the studies indicated that the patterns are similar but not identical.[13]

It has been suggested that changes in the length of rugae with age result from underlying palatal growth.[6,14,15] However, the anterior rugae do not increase in length after 10 years of age according to Van der Linden.[16] Other qualitative characteristics such as shape, direction and unification remain stable throughout life. Despite the ongoing problem of describing palatal rugae patterns qualitatively and quantitatively, their uniqueness to individuals has been recognized in forensic science as providing a potentially reliable source of identification.[9]

Many studies have been carried out on the rugae patterns in the populations of Australia, South Africa and Japan.[17–20] Kapali et al[20] in their study did not reveal any significant differences in the number of primary rugae between Australian Aboriginal males and females. However, Dohke and Osato[19] indicated that among the Japanese, the females had fewer rugae than males. Only one study on palatal rugae pattern in two different populations of India has been performed by Nayak et al,[21] who reported lack of sex dimorphism in their sample.

The purpose of the present investigation was to study the rugae pattern in an Indian male and female sample and to compare the patterns between the two groups, which may assist with differentiating the sexes.

MATERIALS AND METHOD

The study was conducted at the Department of Oral Medicine, Diagnosis & Radiology, Sharad Pawar Dental College & Hospital, Datta Meghe Institute of Medical Sciences University, Sawangi (Meghe), Wardha, Maharashtra, India.

A. Selection of patients

A total number of 120 Class I dentate subjects (n=120), 60 males and 60 females**,** were selected from among the students of the college. All subjects were of Indian origin and between the age group of 22-26 years. All subjects were healthy, free of congenital abnormalities, inflammation, trauma or orthodontic treatment. Subjects were briefed regarding the procedure and written consent was obtained.

B. Impressions

An irreversible hydrocolloid was used as an impression material on an appropriate perforated metal tray for the maxillary dental arch for all subjects. The impressions were then poured with Type III dental stone. All instructions by the manufacturer were followed such as water/powder ratio, vacuum mixing and the use of a vibrator. All casts were free of air bubbles or voids.

C. Method of identification

The method of rugae recording used in this study was based on the classification given by Thomas & Kotze[17] and Kapali et al.[20] These classifications include number, length, shape and unification of rugae. The rugae were highlighted by a black pen marker on the cast under spotlight and a magnification lens. A brass wire was adapted over the rugae and the length of wire recorded using a digital caliper (*Absolute Digimatic, Mitutoyo, Japan)* calibrated to 0.1 mm.

Those rugae which have a length of more than 5 mm are referred to as primary rugae. Secondary rugae are those which have a length between 3-5 mm, whilst fragmentary rugae are those which have a length between 2-3 mm. The shapes of individual rugae were classified into four major types: curved, wavy, straight and circular. Straight types ran directly from their origin to termination. The curved type had a simple crescent shape which curved gently. Evidence of even the slightest bend at the termination or origin of rugae led to a classification as curved. The basic shape of the wavy rugae was serpentine. To be classified as circular, rugae needed to display a definite continuous ring formation. The

direction of each primary rugae was determined by measuring the angle between the line joining its origin and termination and a line perpendicular to the median raphe. Forward-directed rugae were associated with positive angles, backward-directed rugae with negative angles and perpendicular rugae with angles of zero degrees. Unification occurs when two rugae are joined at their origin or termination. Unifications in which two rugae began from the same origin medially but immediately diverged laterally were classified as 'diverging'. Rugae with different origins medially which joined on their lateral portions were classified as 'converging'.

In this study, the secondary and fragmentary types of rugae were ignored when the median value of the total number of rugae was calculated. All the identification and measurements were done by one examiner. To assess intra-observer variation in interpretation, the readings were determined twice for 20 subjects. The data thus obtained was organized and prepared for statistical analysis.

D. Statistical Analysis

Chi-Square test was used for comparison of medians and relationship between the attributes. A significance level of 5% was considered as critical value. In order to calculate the accuracy of sex allocation using rugae shapes a logistic regression analysis (LRA) was performed with sex (where Male=1 and Female=0) as dependent variable and rugae shape as independent variable. Software used was SPSS 10.0 statistical program (SPSS Inc., Chicago, Illinois, U.S.A.).[22] The fit of the logistic model to the data was assessed by an accuracy of fit statistic represented by the -2 log likelihood (-2LL).

RESULTS

The intra-observer error was found to be negligible since the percentage concordance between repeat observations was found to exceed 95 percent with very few discrepancies involving the exclusion of secondary and fragmentary rugae, perhaps because of their size.

Table 1: *Total number of subjects and the mean value of rugae in males and females.*

Sex	Total number of subjects	Total number of rugae*	Mean	SD
Male	60	438	7.30	0.94
Female	60	435	7.25	0.93
z-test	0.29			
p-value	0.77			

** Secondary and Fragmentary types were excluded.*

The total number of rugae and the mean value for males and females is illustrated in Table 1. The distribution of different types of rugae as well as the descriptive statistics is shown in Table 2.

Table 2: *Descriptive statistics of different types of rugae categorized by sex.*

Type of rugae	Sex	No.	Median	z-value
Diverging pattern	M	60	0	3.80*
	F	60	0	
Converging pattern	M	60	0	19.80*
	F	60	2	
Wavy pattern	M	60	3	4.82*
	F	60	2	
Curved pattern	M	60	2	3.82*
	F	60	2	
Straight pattern	M	60	1	0.17
	F	60	1	
Circular pattern	M	60	1	15.33*
	F	60	0	

** Significantly different at the p<0.05 level.*

The wavy and curved pattern of rugae were found to be statistically different in the sexes but were more common in both males and females. The diverging type also showed statistically significant sexual dimorphism but was the least commonly found in both the sexes. However there was a significant sex difference in the converging type, which was found to be higher in females. There was also a significant sex difference in the circular type which was higher in males. The number and difference in length of rugae are shown in Tables 3 and 4. The chi-square showed no significant difference in number and length between the two sexes.

Table 3: *Distribution of the different types of rugae length in males and females*.

Sex	From 5-10 mm	More than 10 mm	Total
Male	219(42.60%)	219(42.60%)	514
Female	215(42.49%)	220(43.47%)	506
χ^2-value	0.14		
p-value	0.29		

Table 4: *Sex differences in number of different types of rugae length.*

Type of rugae	Sex	Median	p-value
From 5-10 mm	M	4	0.59
	F	3	
More than 10 mm	M	4	0.90
	F	4	

The accuracy of sex prediction by LRA is depicted in the classification table (Table 5). Application of LRA to all the rugae types yielded a correct sex allocation rate of 99.2%. This may indicate a high power of sex allocation using the rugae shapes. Table 6 shows the accuracy of the statistic: the lower the −2LL statistic, the better the fit of the model to the data.[22] The −2LL for logistic regression model for rugae type was 5.3, indicating high accuracy of fit of the model to the obtained data.

Table 5: *Classification table of Logistic regression analysis (LRA).*

	Male		Female		Σ	
	n	*%*	*n*	*%*	*n*	*%*
Rugae pattern	60/60	100	59/60	98.3	119/120	99.2

Table 6: *Accuracy of fit statistic (-2LL).*

	-2 Log likelihood
Rugae pattern	5.307

DISCUSSION

It is widely acknowledged that there are limitations in identification by fingerprints, dental records and DNA in some forensic situations and the palatal rugae pattern of an individual may be considered as a useful adjunct for identification purposes. The present study was carried out to study the rugae pattern in an Indian sample and to compare the patterns between the males and females, which may be an additional method of differentiating the sexes, especially if other indicators are missing antemortem.

The method used in this study[7,17,20] was found to be the most practical and easiest to apply compared with other methods such as those of Hauser et al[14] and of Reuer.[23]

The present study was cross-sectional in nature and included recording of the rugae pattern of a narrow age group sample of 22-26 years selected amongst the students of the college who were of Indian origin. Hence further studies across a wider sample age group, larger sample size and with a longitudinal approach may be performed amongst the Indians to corroborate our results.

Nayak et al[21] in their study on population differentiation using rugae shape have mentioned that discrete variables such as rugae shape are better suited for the purpose than continuous variables such as rugae length. The present study did not show any significant difference in the length of rugae between the sexes, whereas rugae shape did, implying that discrete variables are also better suited for sex differentiation.

Kapali et al[20] in their study did not reveal any significant differences in the number of primary rugae between Aboriginal males and females. The present study also did not show any significant difference in the number of rugae between the Indian males and females. These results do not conform to the results presented by Dohke and Osato[19] who indicated that among the Japanese, the females had fewer rugae than males. This may be due to the fact that secondary and fragmentary rugae were not included in the present study and it is the secondary rugae that Dohke and Osato considered in their study leading to sex differentiation.

Thomas and Kotze[24] have noted that although primary rugae have been more widely studied than secondary and fragmentary rugae, they do not possess strong discriminatory ability between different human populations. Only the primary rugae were considered in the present study. Hence, further studies involving comparisons of patterns of secondary and fragmentary rugae between the sexes and also between different ethnic groups, rather than primary rugae alone, could be worthwhile.

Acharya et al[22] in their study on odontometric sex assessment have shown the advantages

of using LRA for sex prediction and have proved the use of LRA as a better alternative to discriminant function analysis (DA) in dental sex assessment. Hence LRA was used in the present study to check the ability of palatal rugae pattern in determining sex. LRA enabled highly accurate sex prediction (99.2%) when the rugae shapes were analyzed. This shows that using the observed rugae pattern, sex can be predicted correctly with a probability of >99% which is a very high degree of prediction and sex allocation.[22] The −2LL for logistic regression model for rugae type was 5.3, indicating an accurate fit of the model to the obtained data. This indicates that palatal rugae shapes may be extremely useful to predict sex. The descriptive statistics also showed significant difference among the sexes in shapes of primary rugae in the present sample (Table 2). The presence of the converging type was found to be significantly higher in females while the presence of the circular type was found to be significantly higher in males. This means that the power of sex prediction may be higher when the converging and circular types of rugae are used. The results reveal that rugae patterns have use as a sex indicator when multivariate statistics such as LRA is applied.

However, these results do not conform to the results presented by Nayak et al[21] who studied the palatal rugae pattern in two populations of India and by using step-wise DA reported lack of sex dimorphism in their sample. The differences may be precluded by the small sample size (30: 15 males and 15 females), narrow age group sample and lack of use of LRA and further work on larger samples of the Indian population may be required to validate the findings.

In this study intra-observer error was assessed by repeat observations and was found to be negligible. Also, the errors in length were small because measurements were categorized rather than retaining their quantitative scale.

However, this error rate may be reduced further or completely eliminated by development of an intraoral scanning device to capture palatal rugae pattern, with image transfer directly to a computer, with appropriate software, as is presently available for fingerprints. This would eliminate the manual errors as well as time involved in the process of categorization of rugae pattern samples. With the use of interconnected computer networks it would be possible to store a large amount of data, facilitating quick retrieval of information to assist with faster and more effective sex differentiation and hence identification.

CONCLUSION

Within the limitations of the present study it may be concluded that the rugae pattern (through the use of LRA) may be an additional method of differentiation between the Indian male and female. This may help narrow the field for identification and give results in conjunction with the other methods such as visual, fingerprints, and dental characteristics in forensic sciences. Further research may be indicated with a larger sample size and with a wider age range in order to substantiate the findings of the present study. In addition, examining the rugae patterns, including the primary, secondary and fragmentary rugae in other Indian populations may further corroborate the findings.

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ESTABLISHING THE RELIABILITY OF PALATAL RUGAE PATTERN IN INDIVIDUAL IDENTIFICATION (FOLLOWING ORTHODONTIC TREATMENT)

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ABSTRACT

This study aims to determine the stability of palatal rugae before and after orthodontic treatment. 50 orthodontic cases were selected with pre- and post-treatment casts and 50 casts were randomly selected as variables. Landmarks on the palatal raphe and rugae were marked on the maxillary casts. Points were made on medial and lateral ends of first, second and third rugae. Each cast was photographed, measured and then trimmed leaving only the rugae area of the hard palate. In the pre and post-treatment group, changes in transverse measurements were significantly different for lateral points of first rugae and anteroposterior changes were significant for the distances between first and second rugae. All inter-point measurements of third rugae were stable in post-treatment casts. Thirty blinded examiners compared 50 trimmed pre-orthodontic casts to similarly prepared one hundred casts for possible matches based on pattern of rugae. The percentages of correct matches for examiners had a median of 90%. The matching of pre-operative and post-operative orthodontic casts demonstrated that although some changes do occur in the rugae during orthodontic treatment, the morphology of palatal rugae remains stable throughout life. Hence carefully assessed rugae pattern may have a definite role in forensic identification. Further, points associated with the third palatal ruage were the most immutable over a person's life and hence could be used as a reference to evaluate the changes in teeth positions during orthodontic treatment.

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Keywords: Palatal rugae, stability, forensic identification.

Running title: Establishing Reliability of Palatal Rugae

INTRODUCTION

Transverse palatine folds or palatal rugae (PR), are asymmetrical and irregular elevations of the mucosa in the anterior third of the palate, arranged in a transverse direction from the palatine raphe located in the mid-sagittal plane.[1-3] The rugae play an important role in forensics, as they are protected from trauma and high temperatures due to their internal position in the oral cavity, surrounded and protected by lips, cheeks, tongue, teeth and bone.

Studies undertaken to determine the thermal effects and the decomposition changes on the palatal rugae have concluded that most burn victims with panfacial third degree burns did not sustain any palatal rugae pattern changes, and when changes were noted, they were less pronounced than the generalized body state.[4] Rugae have been used in medico-legal identification processes because their individual morphological characteristics are stable over time.[1-3] Palatal rugae appear during the third month of intrauterine life from the connective tissue covering the palatine process of the maxillary bone. The growth and development is controlled by epithelial-mesenchymal interactions, where specific extracellular matrix molecules are spatiotemporally expressed during development.[5]

Various investigators have implied that palatal rugae are unique to each individual and they can be used successfully in human identification. However, researchers have disagreed as to whether or not legal identification could be based solely on palatal rugae. Controversy also exists about the stability of quantitative and qualitative characteristics of rugae during growth. English et al[2] and Peavy and Kendrick[6] noted that the characteristic pattern of the palatal rugae did not change as a result of growth, remaining stable from the time of development until the oral mucosa degenerated at death. Van der Linden proved that the anterior rugae do not increase in length after 10 years of age.[7] Also

qualitative characteristics such as shape, direction and unification remain stable throughout life.[2] However, Hauser et al have suggested that the mean ruga count changes moderately in adolescence and then increases markedly from the age of 35 to 40 years.[8] In contrast, Lysell considered that the number of ruga decreased from 23 years of age onwards.[9] Some events can contribute to changes in rugae pattern, including trauma, extreme finger sucking in infancy, and persistent pressure with orthodontic treatment and dentures.[9] It has been suggested that changes in the length of rugae with age result from underlying palatal growth.[8-10] Furthermore, Bailey et al, Almeida et al and Abdel-Aziz et al concluded that movement of teeth may change the position of the rugae points.[11-13]

Very few studies have been undertaken to establish the reliability of palatal rugae pattern in individual identification which could play a very important role in forensic sciences. However, rugoscopy can be applicable for forensic identification only when there is available antemortem information for comparison such as dental casts, tracings or digitized rugae patterns. Previous studies may not have considered the effects of growth, extractions, palatal expansion, or some combination of these. The inadvertent use of other features of the cast, such as teeth, edentulous ridge morphology, muscle attachments, vestibular depth, or some combination of these, to aid in the identification, may have influenced their results. Thus the present study will evaluate accuracy of identification by comparing the rugae patterns on pre-operative and post-operative orthodontic casts overcoming these limitations. The purpose of this investigation is to determine if palatal rugae can be relied upon for identification of the individual and whether it can play a definite role in forensic science.

MATERIAL AND METHODS

To determine whether the pattern of the palatal rugae were affected by orthodontic therapy, one hundred and fifty maxillary casts were collected from various private Orthodontist practitioners, out of which 50 orthodontic cases were selected with pre- and post-treatment maxillary dental casts of patients (21 males and 29 females) ranging from 18 to 27 years. All patients had permanent dentitions at the beginning of treatment. 40 patients had extraction of two maxillary premolars and were treated with fixed orthodontic appliances with subsequent retraction of the maxillary anterior teeth into the extraction spaces. 10 patients had no history of extraction and were treated by conventional edgewise techniques. The duration of treatment varied from 8-30 months. All the initial impressions, which were taken from the orthodontic patients, were made from alginate impression materials and the casts were made from hard dental stone at each of the dental clinics and laboratories.

Cast analysis:

Landmarks on the palatal raphe and palatal rugae were marked on the maxillary casts using a 0.3 mm graphite pencil under adequate light and magnification according to the classification given by Kapali et al[14] (Fig.1). Medial and lateral points were marked on medial and lateral ends of first, second and third rugae. Each cast was photographed by a standardized technique using a tripod mounted SLR digital camera (Nikon D90) with uniform settings and measurements were made using the UTHSCSA Image Tool version 2.0 computer program. The median palatal plane was constructed on the median palatal raphe (MPP). Perpendicular distances from MPP to the rugae points were calculated on each cast. Additionally, transverse linear distances between medial points and between lateral points of the right and left rugae of the first, second and third rugae were calculated. Further, the linear antero-posterior (AP) distances between the medial and between lateral points of all these three rugae types were also individually evaluated. These measurements were compared between the initial (pre-treatment casts) and final records (post-treatment casts).

Palatal rugae for forensic identification; collection of data:

Fifty post-orthodontic casts were dispersed with 50 randomly selected casts which acted as variables. All casts were trimmed so that all areas except for the rugae area of the hard palate are removed. All one hundred and fifty maxillary casts were trimmed utilizing modification of a manner originally described by English et al.[2] The posterior of the cast was trimmed perpendicular to the base until the cast measured 2.5 cm from the incisive papilla to the posterior. The remaining borders were trimmed to shape the arch form until the teeth and vestibule areas had been removed (Fig.2). These one hundred and fifty casts were divided into two groups. The first group of pre-orthodontic casts were given a random

number; the second group consisted of post-operative casts and randomly selected casts mixed together and numbered randomly. The case numbers of the pre-operative casts with that of matching post-operative dental casts were recorded but were not revealed to the evaluators. Thirty examiners including 5 oral pathologists, 5 orthodontists and 20 dental students were selected as evaluators who were given 50 pre-orthodontic casts and were asked to compare them to the other one hundred casts for possible matches based on rugae pattern.

The case numbers that matched correctly were recorded. The percentages of correct matches for each examiner were analyzed. Moreover, we focused on the percentages of correct matches for each case and compared the shapes and patterns of those rugae to find the differences between the cases with low percentages and those with high percentages of correct matches.

Statistical analysis:
The correctness of the match for each examiner was calculated as the percentage of correct matches. The collected data were further analysed using paired t-test to detect any significant differences between the pre- and post-treatment records for the three different groups of measurements. Statistical analysis was performed using SPSS software version 16.

RESULTS

To verify an accuracy of identification based on rugae pattern, we performed the examination of comparing rugae patterns to identify the pairs of pre- and post-treated orthodontic casts (Fig.3). The percentages of correct matches for each examiner ranged from 74-98%. The median percentage was 90%, and the interquartile range was 86–93% (Fig.4).

To examine the indication and the limitations of this method, we analyzed the percentages of correct matches for each case which ranged from 73% to 100%. The median percentage was 90%, and the interquartile range was 87–93% (Fig.5). We also investigated the morphological features which affected the accuracy of the identification.

When the pre- and post- transverse linear changes of the rugae points were compared, average change observed over time was statistically significant for the distance between the lateral points of the first rugae (Table 1). The changes observed in the remaining points in the transverse direction were not significantly different between the two groups. Comparison between the pre- and post-treatment changes in the position of palatal rugae points in relation to the median palatal plane also showed a similar result, with significant observable difference regarding lateral points of first left rugae and lateral points of second left rugae (Table 2). When the anteroposterior changes of the rugae points were compared between the pre- and post treatment casts, the average changes were significant for the distances between the lateral points of the first and second left rugae, medial points of first and second right rugae and lateral points of second and third left rugae (Table 3).

DISCUSSION

In the present study, the rugae points were unstable in the transverse direction with respect to the lateral points of the first rugae. These results concurred with Bailey et al[12] who believed that extraction of the first premolars creates a large space for distal retraction of the maxillary anterior teeth, which affect the positions of the lateral points of the first rugae thus changing the transverse distance between them. These features were first noticed, by Peavy and Kendrick,[6,9,12,15] who said "the closer the rugae are to the teeth, the more prone they are to stretch in the direction that their associated teeth move". These findings were also consistent with those of Van der Linden[7] and Almeida et al[11] who also observed the influence of orthodontic treatment on the positions of the lateral rugae points. None of the medial points of the first rugae were affected for the transverse values. This finding is in congruence with previous studies by Housser,[16,17] who concluded that lateral edges of the rugae move forward with the migration of teeth in extraction cases but felt that the medial ruga points were unaffected. There were no significant changes in transverse values for the medial and lateral points of the second and third rugae. This may be due to a decrease in arch circumference, which primarily affects the anterior part of the palate.[12]

It was also found in the present study that anteroposterior changes were significantly different for distances between lateral points of the first and second left rugae, between second and third left rugae and between the medial points of first and second right rugae,

thus suggesting that that space closure has some effect on the stability of the rugae. These findings concur with Bailey et al[12] who concluded that the extraction of two premolars creates space for distal retraction of the maxillary anterior teeth, which could lead to these changes in anteroposterior values.

Another interesting finding which was found in the present study was the stability of all the measurements of third rugae which could be due to their position near the molar region away from the distal retraction of anterior teeth. Therefore, these points might serve as stable references when evaluating orthodontic treatment as they did not depend on retraction of anterior teeth. These results were consistent with those of Schwarze,[18] who advocated the use of posterior medial rugae points in the evaluation of anteroposterior changes of buccal teeth and Paevy and Kendrick[6] who concluded that these rugae were not appreciably altered after orthodontic treatment. It has been said that the more posterior rugae are less susceptible to changes with tooth movement: the third palatal rugae pair in particular being the most stable reference.[9,13]

On matching of preoperative and post operative casts based on palatal rugae patterns, 30 examiners achieved 90% correct matches as the median in the present study and percentage of correct matches ranged from 74-98% which is in congruence with the results of the following published reports. English et al,[2] who examined the accuracy of identification using essentially the same method as used in the current study, reported that the percentage of correct matches was 100% based on four investigators and two teams, the exception being 88% for one investigator. Limsons and Julian,[4] who compared some points of the rugae patterns using computer software, reported that the percentage of correct matches ranged from 92 to 97% based on four computer operators. Maki Ohtani et al[19] explored the accuracy of using the palatal rugae pattern in forensic practice for personal identification in edentulous cases, and suggested 94% correct matches. Furthermore, in the present study, most of the examiners matched the casts with a high accuracy, in spite of variations in their knowledge regarding oral anatomy and in their experience with forensic identification. With regard to the indication and the limitations of this method, the median of the percentages correctly matched for each case was found to be 90%. These results suggest the usefulness and easy reproducibility of comparing palatal rugae patterns for personal identification of cases. In cases where post-mortem dental identification is difficult as in edentulous patients, rugae pattern can be used as a supplement. Limitations of palatoscopy are that post-mortem identification is not possible without antemortem data. Additionally, complex rugae patterns can cause intra and interobserver errors. Kapali et al have stated that dentures, malpositioned teeth and palatal pathologies can cause alterations in rugae patterns.[14] The matching of pre-operative and post-operative orthodontic casts demonstrated that the changes occurring following extractions and tooth movement did not significantly alter the pattern of rugae. Although some changes do occur in the rugae during orthodontic treatment, the morphology of palatal rugae remains stable throughout life. Hence the pattern of palatal rugae is unique to each individual and that it can therefore be used in establishing identity.

CONCLUSION

Orthodontic treatment and tooth movement have a significant effect on the stability of first and second palatal rugae as concluded by previous investigators. The most reliable points which remain stable over a person's life were the medial and lateral third rugae points and these could be used as reference points to evaluate the dental movements. For future studies it would be beneficial to use a larger sample size and wider age range. Although some changes do occur in the rugae during orthodontic treatment the morphology of palatal rugae remains stable throughout life and may be important for identification where there is antemortem information available for comparison.

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LARGE TABLES/FIGURES ON FOLLOWING PAGES

TABLES

Table 1: *Descriptive statistics for the transverse changes between bilateral rugae points for pre- and post-treated orthodontic patients.*

VARIABLE		MEAN	SD	MAXIMUM	MINIMUM	
1st lateral ruga	Pre	233.623	58.151	328	108.44	**P<0.05**
	Post	250.965	42.240	307	151.2	
1st medial ruga	Pre	57.588	27.098	110.07	16.16	P>0.05
	Post	58.556	20.473	88.77	26.83	
2nd lateral ruga	Pre	276.434	39.220	333.49	226.04	P>0.05
	Post	276.296	36.177	330.49	232.31	
2nd medial ruga	Pre	73.533	41.89	153.69	22	P>0.05
	Post	78.59	31.65	146.51	36.89	
3rd lateral ruga	Pre	297.47	69.98	401.17	143.07	P>0.05
	Post	318.71	32.23	383.54	273.09	
3rd medial ruga	Pre	140.18	57.87	214.22	66.31	P>0.05
	Post	149.98	55.83	222.14	75.18	

Values are expressed in pixels. SD= standard deviation.

Table 2 : *Descriptive statistics for the transverse changes between bilateral rugae points in relation to the median palatal plane for pre- and post-treated orthodontic patients.*

VARIABLE		MEAN	SD	MAXIMUM	MINIMUM	
1st lateral left ruga	Pre	120.56	8.85	166.01	106.92	**P<0.05**
	Post	139.51	18.26	150.12	74.73	
1st medial left ruga	Pre	23.70	14.69	58.14	7.07	P>0.05
	Post	26.04	14.02	56.47	11	
1st lateral right ruga	Pre	122.33	13.35	136.53	110.03	P>0.05
	Post	144.52	17.28	155.08	124.58	
1st medial right ruga	Pre	33.49	15.43	50.36	20.1	P>0.05
	Post	24.75	3.05	28.07	22.09	
2nd lateral left ruga	Pre	142.83	24.81	171.42	127.01	**P<0.05**
	Post	131.95	22.82	158.2	116.84	
2nd medial left ruga	Pre	49.05	20.07	64.28	26.31	P>0.05
	Post	46.01	16	62	30	
2nd lateral right ruga	Pre	121.47	34.32	160.05	94.34	P>0.05
	Post	136.7	27.57	168.01	116.07	
2nd medial right ruga	Pre	46.86	46.99	100.3	12	P>0.05
	Post	44.70	40.98	92	20	
3rd lateral left ruga	Pre	177.06	22.43	202.16	159	P>0.05
	Post	157.49	30.29	192.17	136.24	
3rd medial left ruga	Pre	91.46	6.19	98.18	86	P>0.05
	Post	94.13	3.37	98.02	82.02	
3rd lateral right ruga	Pre	169.38	12.99	162.01	122.07	P>0.05
	Post	173.43	15.98	190	158.11	
3rd medial right ruga	Pre	79.38	54.29	130.06	22.09	P>0.05
	Post	75.54	46.44	106.02	22.09	

Values are expressed in pixels. SD= standard deviation.

Table 3 : *Descriptive statistics for the anteroposterior changes between rugae points for pre- and post-treated orthodontic patients.*

VARIABLE		MEAN	SD	MAXIMUM	MINIMUM	
1^{st} -2^{nd} lateral left ruga	Pre	72.42	24.85	103.48	25.5	**P<0.05**
	Post	54.25	22.53	117.38	41.23	
1^{st} -2^{nd} medial left ruga	Pre	60.85	22.41	94.11	23.6	P>0.05
	Post	57.41	18.50	81.86	25.52	
1^{st} -2^{nd} lateral right ruga	Pre	51.57	13.31	59.93	36.22	P>0.05
	Post	52.99	10.21	59.61	41.23	
1^{st} -2^{nd} medial right ruga	Pre	53.14	27.76	75.29	22	**P<0.05**
	Post	71.18	30.77	96.04	36.77	
2^{nd}-3^{rd} lateral left ruga	Pre	80.25	34.52	137.36	27.66	**P<0.05**
	Post	59.70	36.92	151.94	23.32	
2^{nd}-3^{rd} medial left ruga	Pre	66.53	13.95	90.44	42.43	P>0.05
	Post	64.71	10.92	82.61	45.61	
2^{nd}-3^{rd} lateral right ruga	Pre	70.24	31.52	91.76	34.06	P>0.05
	Post	61.69	15.02	71.69	44.41	
2^{nd}-3^{rd} medial right ruga	Pre	72.34	11.58	82.37	59.67	P>0.05
	Post	88.82	36.82	130.3	60	

Values are expressed in pixels. SD= standard deviation.

FIGURES

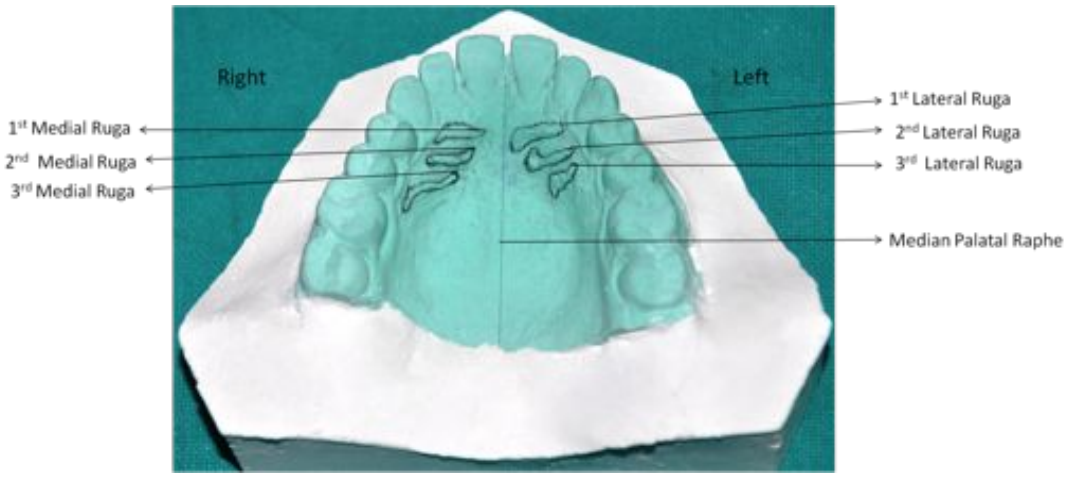

Fig. 1: *Landmarks on a dental cast, showing 1st, 2nd and 3rd rugae with their medial and lateral points.*

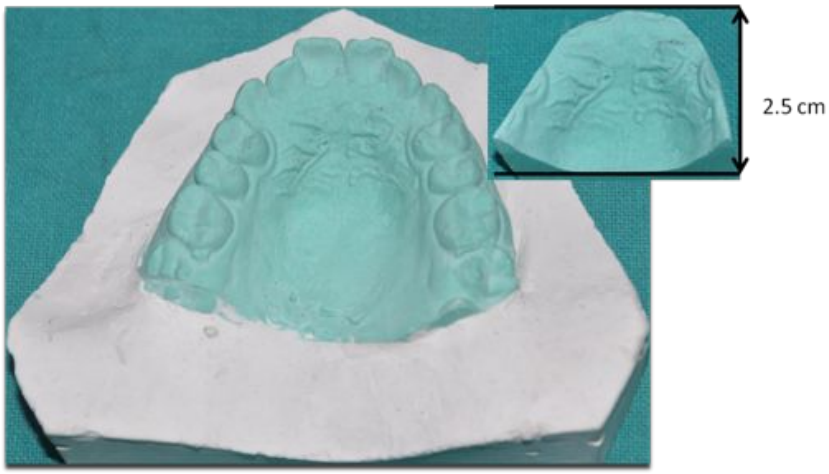

***Fig. 2**: Maxillary cast and trimmed cast. The maxillary casts were trimmed until the casts measured 2.5 cm from the incisive papilla to the posterior, and until the teeth and vestibule region had been removed.*

***Fig. 3:** Matching of pre-operative and post-operative casts based on palatal rugae patterns.*

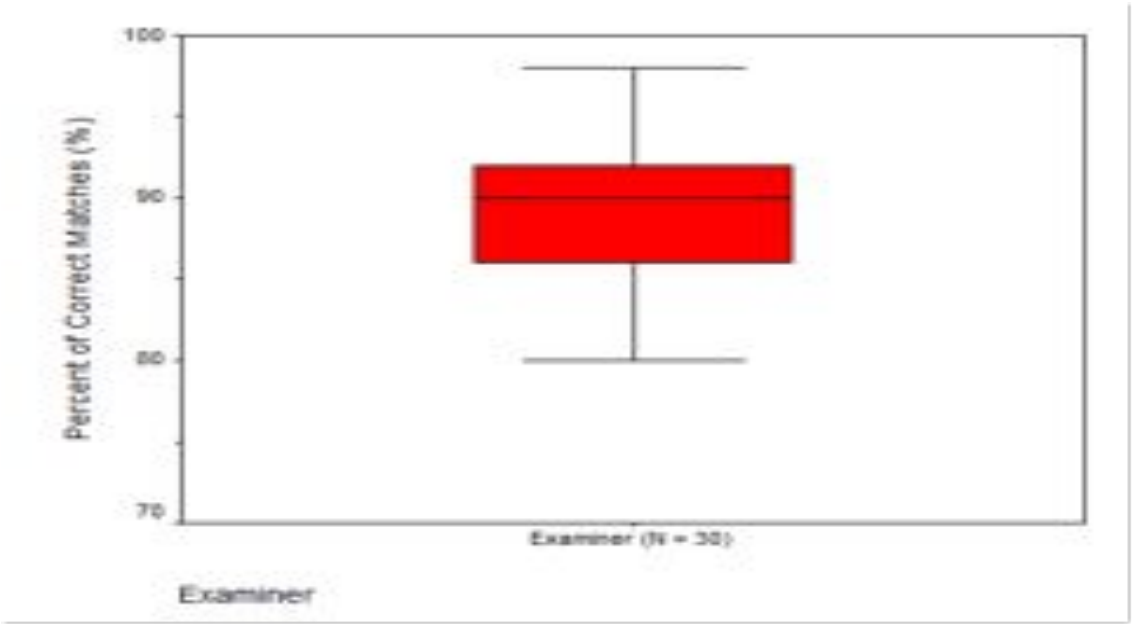

Fig. 4: *Distribution of the percentage of correct matches for each examiner. The median is indicated with horizontal bar, while the horizontal boundaries of the boxes represent the interquartile ranges and the vertical bar indicates the range. The median (interquartile range) of all examiners was 90% (86–93%).*

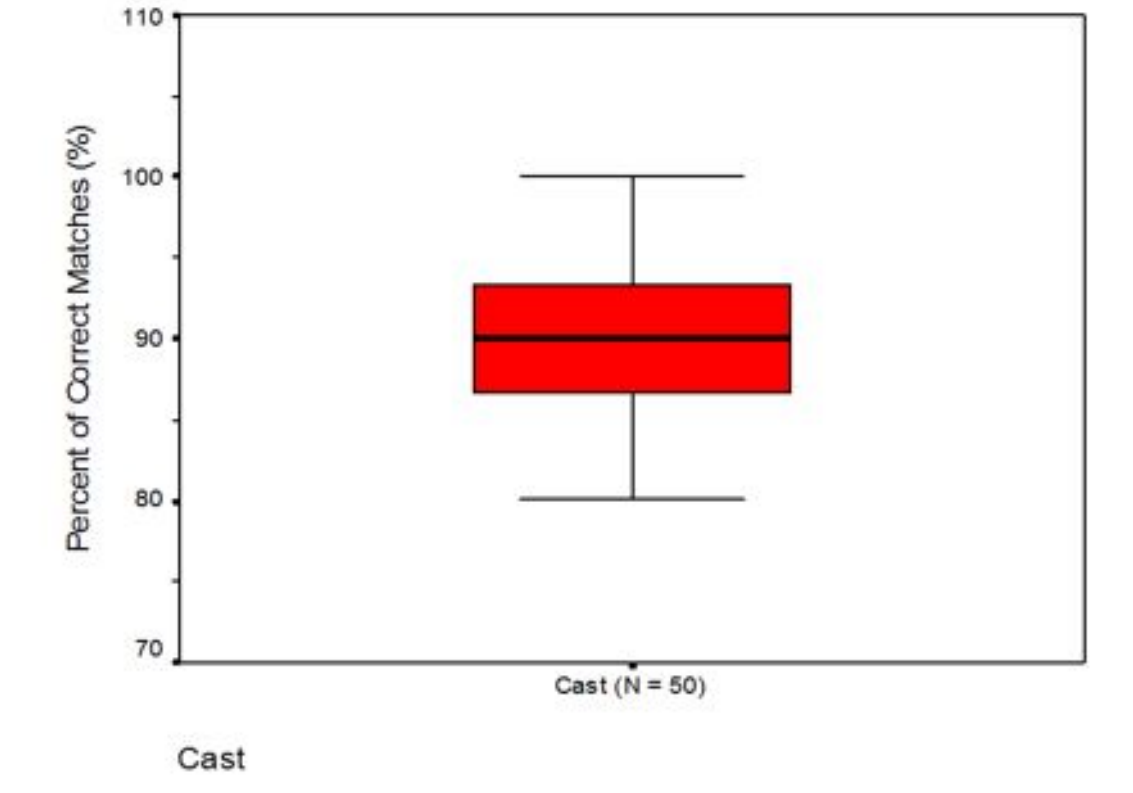

Fig. 5: *Distribution of the percentage of correct matches for each case. The bars and box indicate the same in Fig. 4. The median (interquartile range) of all cases was 90% (87–93%).*

INTERCANINE DISTANCE IN THE ANALYSIS OF BITE MARKS: A COMPARISON OF HUMAN AND DOMESTIC DOG DENTAL ARCHES

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ABSTRACT

One common parameter considered helpful to identify the origin of bite wounds has been the distance between the canine teeth marks left on the victim. The reliability of this parameter to differentiate the origin of the marks (human or animal) was evaluated using a sample of: a) domestic dogs (n=50) weighting between 4.9 kg and 46 kg of undefined breed and b) human beings (n=50). Dog intercanine distances (ID) were measured directly using calipers, those from the human sample were measured from wax imprints using calipers. It was found that dog bite intercanine distance measurements were overall 2.8% wider for the upper arch and 10.4% wider for the lower arch when compared with the overall result for humans. However, it was observed that the measured values for medium sized dogs (between 9.1 kg and 23.0 kg) are similar to the overall results for humans. Therefore, for this range, the stand alone use of intercanine distance measurements from bite wounds marks are inconclusive with respect of defining if of human origin.

(J Forensic Odontostomatol 2011;29:1:30-36)

Keywords: Forensic dentistry; human bites, dogs/AH, identification.

Running title: Bite marks, intercanine distance, human and domestic dog dental arches.

INTRODUCTION

The lesions produced by bites are objects of forensic analysis and one of the first considerations is to determine their origin; whether they were produced by an animal or human. Depending on the outcome, the subsequent investigation will take an entirely different course.

Careful analysis of the dental characteristics and features of a bite mark may help identify whether the biting injury was self-inflicted, caused by an aggressor, an animal or at the very least, may exclude a suspect.[1,2] As soon as a bite mark is detected it should be examined by an expert to determine, among other factors, whether the size and configuration are within human parameters[3] and if there are enough details and preserved remains that reveal any distinguishing features of the dentition.[4,5] Consideration should also be given, for example, to the resistance of the tissue, the anatomical location, position of the bitten person. [6,7]

One of the parameters of the investigation is the measurement of the intercanine distance, as the impressions of the anterior teeth are usually the most evident and likely to be measurable.[8] Spencer[9] highlights the importance of careful measurement of intercanine distances during an investigation. Each injury should therefore be evaluated to determine whether it was produced by human or animal teeth[10] - a distinction that requires comparative knowledge of dental anatomy.[11] The distance between the upper canines in an adult can range from 25 to 40 mm. It has been suggested that a distance less than 25 mm may have been produced by a child,[12] but the recognition of deciduous tooth marks in the bite mark may be a better indicator. For adults in Brazil, the mean intercanine distances in the upper arches has been measured at 29.4 mm, and in the lower arches the mean is 26.7 mm.[5] Bites produced by dogs and other animals often cause much damage with tissue laceration and (avulsion) and human bites can include a wide range of injuries from bruising, abrasions, lacerations and ocassionally tissue avulsion.[13, 14]

The domestic dog (*Canis familiaris*) and humans have a notably distinct morphology of the teeth and arches. This fact may sometimes lead the researcher to believe that the difference between a human bite and one caused by a dog is not at all complex.

However, the movements that take place during the bite, along with particular aspects of the supporting tissue can lead to lesions inflicted by dogs that resemble those caused by humans.

Considering just domestic dogs, the intercanine distance may vary with the animal's breed and weight. This distance in the North American domestic dog ranges from 13.0 to 48.0 mm in the maxilla, while for the mandible there is a range of 6.0 to 49.0 mm.[15] Although there is awareness of this variation, there are no Brazilian studies that present tables of measurements in order to guide the differentiation of bites produced by dogs and those produced by human dentitions.

The head and neck are the most frequent sites of injury in victims bitten by dogs, and bites occasionally result in death. There are very few studies on injuries left by dog bites in humans.[16] However, it is an important topic, due to the fatal attacks by aggressive breeds of dogs; 85% of fatal cases occur in children under 12 years.[17] All types of dogs are capable of inflicting injury on people and the conclusive and objective determination of the breed of dog is only possible by examining the pedigree (potentially time-consuming and complicated) combined with DNA testing. Mixed breed dogs, or those that have not had their pedigree registered cannot be recognised as a certain breed and their description is usually vague and based on subjective visual observation.[18] More than 30 breeds of dogs were described as being responsible for fatal attacks on people in the United States from May 1975 to April 1980 and in many cases they were dogs of mixed origin or unknown breed.[19]

The masticatory force of dogs of different breeds varies with the excitement of the biting animal, and also with its weight.[20] The shape and size of the skull have been suggested as factors related to the variation in the size and position of the teeth.[16,20,21] It is important to consider that there is always the possibility of distortion, hindering or even making it impossible to physically analyse the bite mark[22] and that guidelines laid down by the forensic odontology community for such an analysis should be respected.[23]

Thus, to evaluate whether the intercanine distance is a reliable parameter to differentiate between bite marks produced by humans and domestic dogs, this distance was measured for the upper and lower arches in a sample of 50 domestic dogs of different weights and 50 human subjects. Knowledge of these characteristics may enable greater scientific certainty in establishing the differential diagnosis of bite marks; their study is an important area of forensic odontology and there is a lack of clear parameters for classification.

MATERIALS AND METHODS

The study used no. 7 pink wax plates, a heating lamp, plastic containers and a caliper for both the human and dog sample. The researchers also used personal protective equipment (PPE). A single examiner was responsible for carrying out all measurements and intra-examiner calibration was performed by successive measurements in predetermined scale (millimeter ruler) with no significant differences. The results were also evaluated by a second observer, resulting in agreement between them.

Human sample: The study sample consisted of 50 bite marks made by human subjects, young adults (students of the Faculty of Dentistry, University of São Paulo), of both genders, on plates of pink wax (no. 7). The decision was made not to distinguish this sample by gender or age, because such information is not initially obtained when evidence of bite marks is found. After explanation and justification of the objectives of this study, consent was obtained and consent forms signed, which allowed the use of the material (plates with bite mark impressions and the values obtained for the intercanine distance). The students were asked to bite into a wax plate that was folded in half and slightly softened by the heat of the lamp. The plates with the bite impression were placed in individual plastic envelopes and handled by the researchers, always with the use of personal protective equipment (PPE). Measurements were taken with a caliper, noting the distances in mm between the tips of the right and left canine (as measured in a straight line) imprinted in the wax plate in the maxilla and mandible

separately, and the values were recorded on separate forms (Fig. 1).

Sample of domestic dogs: (*Canis familiaris*): The study sample consisted of 50 dogs with no restriction concerning breed, size or weight; they were attended to at the Laboratory of Comparative Dentistry, Faculty of Veterinary Medicine and Animal Science, University of São Paulo. With permission granted by those responsible for the dogs, measurements of the domestic dogs' intercanine distances were taken directly from the mouth using a caliper. During the study the animals were sedated, in preparation for scheduled procedures that were separate to our research. To calculate the amount of sedation required, animals were weighed beforehand and these data were also recorded. There was no removal of biological material, use of drugs or exposure to stress, pain, restriction of water or starvation due to this study, in accordance with ethical principles involving research with animals.

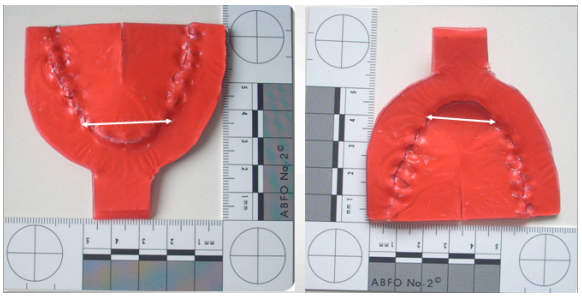

Fig. 1: *Human sample: measuring the distances in mm between the tips of the right and left canine imprinted in the wax plate, in the maxilla and mandible.*

The distinction by breed of dogs was not considered since the determination of a specific breed requires reliable methods that are not available to the general population. The animals in this sample were considered "no defined breed" (NDB). Due to the extensive range of sizes of the domestic dog found in nature or due to intervention by man in the selection of breeding, animals were divided according to their physical size, taking into consideration their weight in kilograms: small size (for animals with weight equal to or less than 9.0 kg), medium (for animals over 9.1 kg up to 23.0 kg), large size (for those weighing between 23.1 kg and 40.0 kg), and giant-sized (for animals over 40.1 kg), according to the classification proposed by Goldston and Hoskins.[24] The measurements in millimeters of the distance between the cuspid tips of the right and left canine were considered, and were taken from the upper and lower arch using a caliper (Fig. 2).

Because this evaluation of the measurement of intercanine distances from humans and domestic dogs only used a caliper, there was no risk to the participants and the confidentiality of identity was preserved during the research. For the descriptive statistics of the sample, the statistical package BioEstat 4.0[25] was used, with a confidence level of 95%.

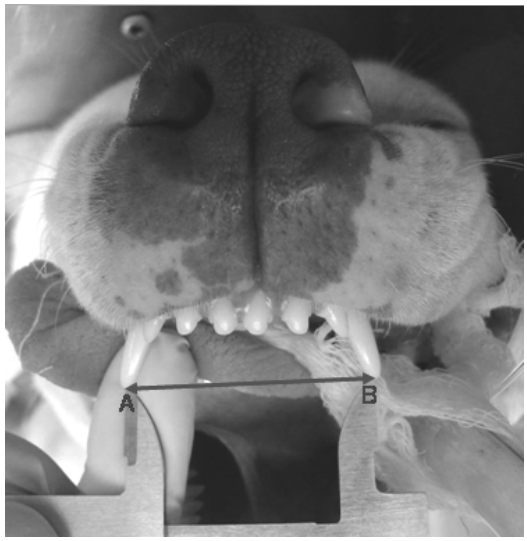

Fig. 2: *Domestic dog upper arch intercanine distance measurement (A to B).*

RESULTS

For the 50 dogs, the mean weight was 14.3 kg, with a standard deviation of 8.2. The lightest was 4.9 kg and the heaviest was 46.0 kg. In order to analyze the measurements of intercanine distances of the animals they were divided into four levels according to their physical size relative to body weight in kilograms: less than or equal to 9.0 kg (small), from 9.1 kg to 23.0 kg (medium), from 23.1 kg to 40.0 kg (large) and greater than 40.1 kg (giant). Fifty-eight percent (n = 29) of the animals were medium sized, with a mean weight of 13.8 kg, 28% (n = 14) were small with a mean weight of 7.3kg, 12% (n = 6) were large size, with a mean of 27.6 kg and only 2% (n = 1) were giant-sized, 46.0 kg.

Regarding the intercanine distance, the mean for the maxilla for the whole animal sample (n = 50) was 35.3 mm, with a standard deviation of 7.9 and for the mandible it was 30.6 mm, with a standard deviation of 5.6. The lowest values found were 22.0 mm in the maxilla and 18.0 mm in the mandible (of the same dog), which weighed 6.1 kg (small). The highest values were 65.3 mm in the maxilla and 45.1 mm in the mandible (of the same dog), which weighed 46.0Kg (giant). The dog sample with the lowest weight (4.9 kg) had an intercanine distance of 30.0 mm in the maxilla and 25.4 mm in the mandible.

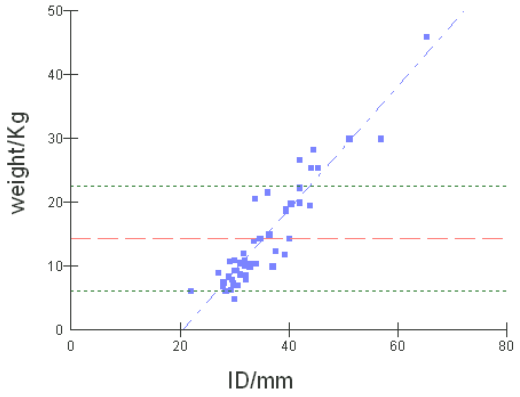

Fig. 3: *Correlation between body weight (dog) and intercanine distance in the maxilla.*

When we consider the stratification of the sample per weight, the means that were found for the intercanine distance in the maxilla and mandible, respectively, were 29.0 mm and 25.6 mm for small dogs, 34.9 mm and 30.7 mm for medium dogs, 47.2 mm and 39.7 mm for large dogs, and 65.3 mm and 45.1 mm for giant dogs (Table 1).

Table 1: *Distribution of the sample of dogs according to the frequency, percentage and means of weight and intercanine distances in the maxilla and mandible.*

Dogs				Means		
Size	Weight/ kg	#	%	Weight / kg	Intercanine distance / mm	
					maxilla	mandible
Small	<9.0	14	28	7.3	29.0	25.5
Med	>9.1-23.0	29	58	13.8	34.9	30.7
Large	>23.1-40.0	6	12	27.6	47.2	39.7
Giant	>40.1	1	2	46	65.3	45.1

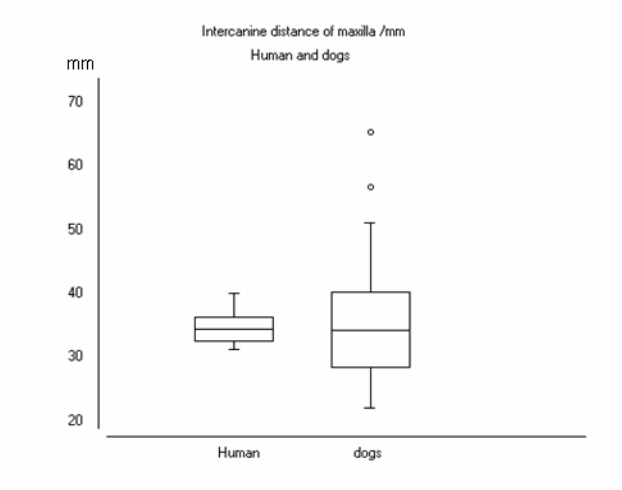

Fig. 4: *Intercanine distance of maxilla/mm of the sample (human and dogs).*

A correlation (*Pearson*) was found between the animal's weight and the intercanine distance (ID) for both the maxilla (r = 0.93) and the mandible (r = 0.83). Figure 3 shows this correlation between weight and maxillary intercanine distance. The descriptive statistical analysis for the human sample revealed that the mean intercanine distance for the maxilla in the entire sample (n = 50) was 34.3 mm with a standard deviation of 1.8 and in the mandible this mean was 27.5 mm with a standard deviation of 1.7. The lowest values were 31.2 mm in the maxilla and 25.0 mm in the mandible.

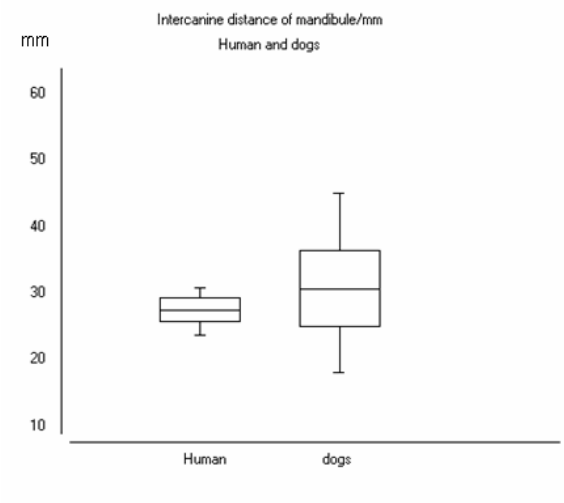

Fig. 5: *Intercanine distance of mandible/mm of the sample (human and dogs).*

Figures 4 and 5 present the values for intercanine distances in the maxilla and mandible of the entire sample (human and dogs) and it is possible to see that the measurements are concentrated between the values of 25 and 35 mm. The differences between the mean IDs in the maxilla and the mandible found in human subjects and dogs varied with the size of the animal; the smallest difference was found in the comparison with medium-sized animals. For animals of this size, distances were 1.7% higher in the maxilla and 10.4% in the mandible (Table 2). The ID results for the sample of dogs (n = 50) suggest a difference of 2.8% higher for the upper arch and 10.4% for the lower arch when compared with human intercanine distances (a reference mean in this study of 34.3 mm for the maxilla and 27.5 mm in the mandible).

Table 2: *Difference in mean intercanine distances in the maxilla and mandible between medium-sized dogs and humans.*

	Medium-sized dogs	Humans	Difference means / mm	% ≠
ID maxilla / mm	34.9	34.3	0.6	1.7
ID mandible / mm	30.7	27.5	3.2	10.4

DISCUSSION

Forensic literature has shown that both domestic dogs and humans can produce bite wounds and the identification of the aggressor is the determining factor in subsequent investigations. In order to direct the investigations into bite marks one attempts to observe if the dimensions and configuration of the lesion allow an identification of whether they were produced by humans or animals, and the intercanine distance is one of the parameters that is used by several authors.[2,3,5,7-9,11,12,14,16] It is important to note that humans have four incisors per dental arch, while dogs have six. However, the marks left behind do not always show the full arch and the distortions produced by the elasticity and retractility of tissues, movement and amount of contact can lead to misinterpretation.

If one only considers the morphology and anatomy of the teeth of dogs and humans, there would certainly be no difficulty in differentiating the markings produced by the two species as distinct. But given the dynamics imposed during the biting act and the reaction of the victim, what is observed is not a simple impression of teeth on a substrate. Where the biting injury quality allows the identification of puncturing lesions, suggestive of penetration of canine teeth, the distance between these marks is measured in an attempt to work out to which species they belong.

The results of this study show a correlation between the intercanine distance and weight of domestic dogs and a greater weight implies a larger animal. However, the sample size did not allow us to indicate the existence of maximum and minimum values, especially because the variance was significant. The measurements taken in this human sample are within the range published by the American Academy of Pediatrics[12] and the mean for the maxilla was 14% and for the mandible 2.9% higher than those found by Marques et al[5] also in a sample of Brazilians. However the human sample in our study was not standardized. It is noteworthy, however, that for medium-sized animals (over 9.1 kg and up to 23.0 kg), we obtained values closest to those in the human sample, with values that were only 1.7% greater for the maxilla. The analysis of these distances alone

would not differentiate whether the offending agent was animal or human.

CONCLUSIONS

The intercanine distance of both the maxilla and the mandible in dogs is related to the animal's weight, however, the size and shape of skull related to the breed of the animal must be considered in future studies. The variability of the intercanine measurements found in both humans and domestic dogs had similar values, but on average measurements for the dogs are larger. Intercanine distances measuring between 25.0mm to 35.0mm were found in both humans and different sizes of the canine species. These differences are also suggestive of significant individual variation, which may help identify the biter, and distinguish the impression left by the bite. We understand that the intercanine distance when found and measured in bite marks (on its own) does not allow a conclusive analysis in determining the origin: animal or human, especially when measurements for medium sized dogs, as in this study, are similar to the overall results for humans. Therefore, further studies should be carried out, in an attempt to clarify the origin and differentiation of biting injuries.

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SEXUAL DIMORPHISM IN THE PERMANENT MAXILLARY FIRST MOLAR: A STUDY OF THE HARYANA POPULATION (INDIA)

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ABSTRACT

Sexual identification of immature skeletal remains is still a difficult problem to solve in Forensic Anthropology. The aim was to evaluate the existence of sexual dimorphism in maxillary first molars. The base sample comprised 200 subjects (100 males and 100 females) aged 17–25 years. The buccolingual (BL) and mesiodistal (MD) diameters of maxillary first molars were measured using digital vernier calipers both intraorally and on study casts. Data was analyzed using Independent sample t-test and paired t-test. Results showed statistically significant sexual dimorphisms in male and female odontometric features. The mean values of the parameters were greater on the left side than on the right side. Amongst the intraoral group, the right maxillary first molar was found to exhibit the greatest sexual dimorphism (5.34%) in terms of buccolingual dimension. Amongst the study cast group, the left maxillary first molar was found to exhibit the greatest sexual dimorphism (5.54%) in terms of buccolingual dimension. The buccolingual dimensions exhibited greater sexual dimorphism than mesiodistal dimensions. Conclusion: sex determination from an incomplete skeleton or young children may be difficult and in such situations the odontometric features of the teeth can be of immense help in determining the sex.

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Keywords: Maxillary first molar, buccolingual (BL), mesiodistal (MD), sexual dimorphism

Running title: Sexual variation in permanent maxillary first molar

INTRODUCTION

Sex determination of skeletal remains forms part of archaeological and medicolegal examinations. The method may vary and depend upon the available bones and their condition.[1] The identification of sex is of significance in cases of mass fatality incidents where bodies are damaged beyond recognition.[2] Further, in situations where only fragments of jaw bones with teeth (or teeth alone) are found, then sex determination is possible only with the help of teeth.

Teeth, being the hardest and chemically the most stable tissue in the body are an excellent material in living and non-living populations for anthropological, genetic, odontologic and forensic investigations.[3] Tooth size standards based on odontometric investigations can be used in age and sex determination.[4] With such tooth size standards, whenever it is possible to predict the sex, identification is simplified because then only missing persons of one sex need to be considered. In this sense identification of sex takes precedence over age.[5]

Sex determination using dental features is primarily based upon the comparison of tooth dimensions in males and females or upon the comparison of frequencies of non-metric dental traits like Carabelli's trait of upper molars, deflecting wrinkle of the lower first molars, distal accessory ridge of the upper and lower canines or shovelling of the upper central incisors.[6] This is based on the fact that although the morphology of the tooth structure is similar in males and females, the size of the tooth does not necessarily remain the same, as the tooth size is determined by cultural, environmental, racial and genetic factors.[7] 'Sexual dimorphism' refers to those differences in size, stature and appearance between male and female that can be applied to dental identification because no two mouths are alike.[8] Many studies have established that amongst all the teeth, the permanent mandibular canines are found to exhibit the greatest sexual dimorphism.[9]

The purpose of this study was to evaluate the existence of sexual dimorphism using both buccolingual and mesiodistal dimensions of maxillary first molars, as they are the first permanent teeth to erupt into the oral cavity at the mean age of 6-7 years and are less commonly impacted as compared to canines. Further, sex can be determined accurately in mature individuals if the postcranial skeleton is intact. But in young children, determination of sex from the skeleton is difficult.[10] In such situations, the odontometric features of teeth that erupted at an early age can be of immense help in determining the sex.

MATERIAL AND METHODS

The base sample comprised 200 students (100 males and 100 females) of an age group ranging from 17-25 years, selected from M.M College of Dental Sciences and Research, Mullana, Ambala, Haryana, India. This particular age group was studied as ante-mortem insults such as attrition and abrasion affecting occlusal and approximal tooth surfaces are minimal. The inclusion criteria taken into consideration were as follows:

- Healthy state of periodontium
- Caries free teeth
- Presence of bilateral maxillary first molars

Following informed consent, impressions of the maxillary arch were made with irreversible hydrocolloid (alginate) material and casts poured immediately in type II dental stone to minimize dimensional change. The buccolingual (BL) and mesiodistal (MD) diameters of the maxillary first molars were measured using digital vernier calipers (resolution 0.01mm) both intraorally and on study casts.

MD diameter of the crown: This measurement is the greatest mesiodistal dimension between the contact points of teeth on either side of jaw.

BL diameter of the crown: This measurement is the greatest distance between buccal and lingual surfaces of the crown, taken at right angles to the plane in which the mesiodistal diameter is taken.

The measurements were performed by one person and all values were rounded to two decimal places. In order to assess the reliability of the measurements, intra-observer error was tested. The same measurements were obtained from 100 randomly selected teeth from the original sample at a different time by the same author to assess intra-observer error. Another observer measured the same randomly selected teeth in order to test the inter-observer error. Their measurements were analyzed using Student's t-test. There was no statistically significant difference between the findings of the two observers.

Statistically significant sexual dimorphisms in male and female odontometric features were tested by the unpaired t-test. The differences in the mean values of the parameters between the right and the left side measured intraorally and on study casts were tested using the paired t-test. The level of statistical significance was set at $p < 0.05$.

The mean values of BL and MD dimensions of males and females were subjected to the formula[11] to calculate sexual dimorphism

$$\text{Sexual dimorphism} = \frac{X_m - 1 \times 100}{X_f}$$

Where X_m = mean value for males and
X_f = mean value for females

OBSERVATIONS AND RESULTS

The following parameters were determined intraorally and on the study casts in males and females:

a) BL diameter of right and left maxillary first molars

b) MD diameter of right and left maxillary first molars

- ✓ It was observed that the comparison of mean values of parameters showed highly statistically significant differences between males and females, with $p<0.001$; measured both intraorally (Table 1) and on the study casts (Table 2).
- ✓ The mean values of the parameters were greater on the left side than on the right side whether measured intraorally or on the study casts.
- ✓ The paired t-test showed that the differences in the mean values of the parameters between the right and the left side were statistically significant with respect to pairs 1, 2 and 4 with

p value<0.05; while with respect to pair 3, the difference was highly statistically significant with p value<0.001 (Table 3).

- ✓ The mean values of the parameters in both males and females were greater on the left side than on the right side. The paired t-test showed that the differences in the mean values of the parameters with respect to pairs 2 and 3 (in males); pairs 3 and 4 (in females) were highly statistically significant with p value<0.001. The difference was statistically non-significant with respect to pairs 1 and 4 (in males), and pair 2 (in females) with p value>0.05. While the difference was statistically significant with respect to pair 1 (in females) with p value<0.05 (Table 4).
- ✓ Sexual dimorphism amounted to 5.34% and 5.16% for right and left buccolingual dimensions of maxillary first molars respectively as compared to 4.51% and 4.55% for right and left mesiodistal dimensions of the same teeth measured intraorally (Table 5).
- ✓ Sexual dimorphism amounted to 5.44% and 5.54% for right and left buccolingual dimensions of maxillary first molars respectively as compared to 4.74% and 4.84% for right and left mesiodistal dimensions of the same teeth measured on study casts (Table 5).
- ✓ Amongst the intraoral group, the right maxillary first molar was found to exhibit the greatest sexual dimorphism (5.34%) in terms of buccolingual dimension while it is the least dimorphic (4.51%) in terms of mesiodistal dimensions.
- ✓ Amongst the study cast group, the left maxillary first molar was found to exhibit the greatest sexual dimorphism (5.54%) in terms of buccolingual dimension while the least dimorphic value was that for right maxillary first molar (4.74%) in terms of mesiodistal dimensions.
- ✓ Comparing both the groups, all the measurements of maxillary first molars on the left side exhibited greater sexual dimorphism than their respective counterparts, except for the right buccolingual measurement taken intraorally.
- ✓ Comparing the linear measurements, the buccolingual dimensions of maxillary first molars were found to exhibit greater sexual dimorphism than mesiodistal dimensions of the same teeth.

Table 1: *Comparison of mean values of different parameters in males and females measured intraorally using unpaired t-test (right and left maxillary molars).*

Parameters	Sex	Mean (mm) ± S.D	p value
BL-R	M	11.09 ± 0.35	<.0001
	F	10.53 ± 0.42	
BL-L	M	11.10 ± 0.36	<.0001
	F	10.56 ± 0.43	
MD-R	M	10.51 ± 0.44	<.0001
	F	10.06 ± 0.31	
MD-L	M	10.52 ± 0.44	<.0001
	F	10.06 ± 0.31	

Table 2: *Comparison of mean values of different parameters in males and females measured on study casts using unpaired t-test (right and left maxillary molars).*

Parameters	Sex	Mean (mm) ± S.D	p value
BL-R	M	10.93 ± 0.36	<.0001
	F	10.37 ± 0.42	
BL-L	M	10.96 ± 0.36	<.0001
	F	10.38 ± 0.42	
MD-R	M	10.37 ± 0.44	<.0001
	F	9.90 ± 0.31	
MD-L	M	10.39 ± 0.45	<.0001
	F	9.91 ± 0.31	

Table 3: *Comparison of differences in the mean values of parameters between the right and the left side measured intraorally and on the study casts using paired t-test.*

Pairs	Parameters	Mean (mm) ± S.D	95% Confidence interval of the difference		p value
			Lower	Upper	
Pair 1	Intaoral BL-R - BL-L	-0.02 ± 0.12	-0.039	-0.005	.009
Pair 2	Intraoral MD-R - MD-L	-0.006 ± 0.03	-0.011	-0.001	.008
Pair 3	Study cast BL-R - BL-L	-0.017 ± 0.02	-0.020	-0.013	.000
Pair 4	Study cast MD-R - MD-L	-0.013 ± 0.07	-0.023	-0.003	.009

Table 4: *Comparison of differences in the mean values of the parameters in males and females measured both intraorally and on study casts using paired t-test.*

Sex	Pairs	Parameters/ Group	Mean (mm) ± S.D	95% Confidence interval of the difference		p value
				Lower	Upper	
M	Pair 1	Intraoral BL(R) - BL(L)	-0.013 ± 0.10	-0.03	0.006	0.187
	Pair 2	Intra oral MD(R) – MD(L)	-0.008 ± 0.02	-0.01	-0.003	.0001
	Pair 3	Study cast BL(R) – BL(L)	-0.022 ± 0.02	-0.02	-0.017	.0001
	Pair 4	Study cast MD(R)–MD(L)	-0.018 ± 0.09	-0.03	0.001	0.069
F	Pair 1	Intra oral BL(R)- BL(L)	-0.031 ± 0.13	-0.05	-0.004	0.024
	Pair 2	Intra oral MD(R) – MD(L)	-0.004 ± 0.04	-0.01	0.003	0.278
	Pair 3	Study cast BL(R) - BL(L)	-0.011 ± 0.01	-0.01	-0.007	.0001
	Pair 4	Study cast MD(R) – MD(L)	-0.086 ± 0.02	-0.01	-0.004	.0001

Table 5: *Percentage Sexual Dimorphism in Maxillary first molars.*

GROUP	BL-R	BL-L	MD-R	MD-L
Intraoral	5.34%	5.16%	4.51%	4.55%
Study Cast	5.44%	5.54%	4.74%	4.84%

DISCUSSION

Sex determination is one of the prime factors employed to assist with the identification of an individual. The accuracy of sex determination using diverse parameters of the body such as craniofacial morphology and measurements on the pubis ranges from 96% to 100%.[12,13] Correct sex identification limits the pool of missing persons to just one half of the population. In forensic contexts, however, it is not uncommon to recover partial remains, with fragmentary skull and pelvic bones. The teeth are one of the strongest human tissues and are known to resist a variety of ante-mortem and post-mortem insults.[14]

The human dentition has a complement of 32 teeth; at least a few teeth may be recovered. Hence, they are routinely used in comparative identification of human remains. The fact that most teeth complete development before skeletal maturation makes the dentition a valuable sex indicator, particularly in young individuals.[15]

The present study established the impact of sex factor on the morphometry of maxillary first molars. Buccolingual and mesiodistal diameters of right and left maxillary first molars in males and females were measured both intraorally and on study casts. The comparison of mean values of parameters measured between males and females showed highly statistically significant differences with $p<0.001$and these results were in agreement with the studies done by Perzigian AJ[16], Ghose LJ et al.[17], Stroud JL et al.[18], Hattab FN et al.[19], Rai B et al.[20] and Ghodosi A et al.[21], in which they have observed that the males had larger teeth than females in all the dimensions. Such differences in dimensions of the teeth can be due to greater dentine thickness in males as compared to females, as the Y chromosome increases the mitotic potential of the tooth germ and induces dentinogenesis; whilst the X chromosome induces amelogenesis.[6]

The present study showed that the mean values of all parameters were greater on the left side as compared to the right side whether measured intraorally or on study casts. The results were in agreement to the study done by Rai B et al.[20], who found the left buccolingual dimensions of maxillary first molars to be greater than its counterparts. Similar results were obtained by a study done by Zarringhalam M,[22] who found that dimensions of all permanent teeth were greater on the left side than the right side in upper jaw while it was reverse in the lower jaw. Right-Left differences may be attributed to dental asymmetry; as perfectly bilateral body symmetry is a theoretical concept that seldom exists in the living organisms.

Amongst the intraoral group, the right maxillary first molar was found to exhibit the greatest sexual dimorphism (5.34%) in terms of buccolingual dimension. The results of the present study were in agreement with the study done by Rai B et al,[20] who found a similar result with the right maxillary first molar exhibiting the greatest sexual dimorphism (8.9%). Amongst the study cast group, the left maxillary first molar was found to exhibit the greatest sexual dimorphism (5.54%) in terms of buccolingual dimension while the least dimorphic value was that for the right maxillary first molar (4.74%) in terms of mesiodistal dimensions.

Comparing the linear measurements, the buccolingual dimensions of maxillary first molars in the present study were found to exhibit greater sexual dimorphism than mesiodistal dimensions of the same teeth. The results of this study are in agreement with the study done by Garn SM et al,[23] who found that among 117 adolescents, sexual dimorphism amounted to 5.6% for the buccolingual diameter as compared to 4.2% for the mesiodistal diameter of the same teeth.

The method in the present study employed is simple and inexpensive to conduct and therefore can be applied in forensic odontology for establishing sex identity of an individual.

SUMMARY AND CONCLUSION

The emerging field of forensic odontology in India relies a lot on inexpensive and easy means of identification of persons from fragmented jaws and dental remains. It is in such situations that the dentist can be called upon to render expertise in forensic science. A database may be established of dental morphometric measurements of non-attrited teeth with a view to determining the variations amongst large populations that may be beneficial for anthropological, genetic, legal and forensic applications.

The present study established the existence of statistically significant sexual dimorphism in maxillary first molars. But in order to raise the level of confidence and percentage in success of determining sex, it is best to combine several different methods, when possible, especially when the ante-mortem data on sex are not available (most commonly in archaeological series).

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CASE REPORTS AND BACKGROUND: DIFFICULTIES WITH IDENTIFICATION - SWEDEN

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ABSTRACT

Despite the best conditions such as professional management, all possible aid and means of assistance, along with good legislation, sometimes unexpected factors occur to prevent or at the very least delay identification of the unknown deceased. The specific difficulties in identification cases that involve several countries, as well as problems arising from inconsistencies created in the *Antemortem* (AM) and *Postmortem* (PM) files, can obstruct the identification of the recovered human remains. There may be long delays with police procedures whenever a missing person or a dead body has crossed a national border. Also, lack of professional dental investigation can make comparisons difficult or sometimes impossible. Three cases from Swedish files have been used to illustrate such difficulties - there were some parts of the investigations that worked better than others as well as specific problems that arose from the mistakes and delays that occurred. Improving standards and learning from such difficulties may help to minimise future problems.

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Keywords: Forensic odontology, identification, dental record, DVI System International

Running title: Difficulties with identification cases

INTRODUCTION

The aim of this paper is to give a general background to dental identification in Sweden and describe three cases to illustrate some of the difficulties encountered with identification.

The Swedish National Board of Forensic Medicine is a government agency that comprises forensic medicine and forensic odontology, forensic psychiatry, forensic toxicology and forensic genetics. The National Board of Forensic Medicine is separated from the police forces in order to be able to keep its neutrality and objectivity while handling the forensic parts of any police investigation.

Around 250 single case identifications are made by forensic odontologists in Sweden every year. The majority of these cases (about 70 %) are carried out at the forensic medicine department in Stockholm which is responsible for the most densely populated area of the country: around two million people. Sweden has a population of nine million mostly living in urban areas of the southern part of the country. There is one full time forensic odontologist position in Sweden, placed at the forensic medicine department in Stockholm.

The forensic odontologist is responsible for the identification of single cases at the forensic medicine departments in Stockholm and Uppsala, keeping the repository of missing persons and unidentified bodies of the entire country, as well as taking care of other forensic odontology issues, for example: age estimations, bite mark and dental trauma analyses in criminal investigations (not insurance or malpractice cases). All education and research in forensic odontology, as well as keeping up to date on the legislation that governs the dental care and its documentation in Sweden, is also a part of the everyday work of the forensic odontologist. The forensic odontologist is also a natural member of the Swedish Identification Commission, which is activated whenever there is a fatal disaster abroad involving Swedish citizens. Apart from the employed forensic odontologist there are a few dentists contracted by the National Board of Forensic Medicine to carry out identifications at four other forensic medicine departments in Sweden; two of these dentists are specialists in oral radiology and the other two are forensic pathologists as well as dentists.

Most of the identification cases in Sweden are relatively straightforward, often requiring only confirmation of the identity of a deceased individual, because the police can provide a tentative identity. These cases mostly involve people who have died at home and have not been found for extended periods of time, fire victims and transport fatalities. The police will provide the dental records from the victim's dentist for comparison with the dental PM findings and radiographs. However, every now and then a totally unknown body is found.

Sweden is a big, sparsely populated country with thousands of square miles of forests and thousands of lakes and a very long coastline. On top of that Sweden shares the Baltic Sea with nine other countries. The water is cold

most of the year and the ice can be treacherous, especially in the spring. Whenever human remains are found outdoors, or in the water, the repository of missing persons becomes of importance. Sweden, together with other Nordic countries, has a very good record of identifying totally unknown bodies; only one or two persons are buried as unknown each year. This is partly due to good cooperation between the police force and the forensic odontologist, as well as the good collaboration between the countries around the Baltic Sea. Despite everything that facilitates the work of the forensic odontologist there are some cases that have proved to be more challenging, demanding long periods of research before achieving the desired result - an identification.

THE REPOSITORY OF MISSING PERSONS AND UNIDENTIFIED BODIES

More than 7000 persons are reported missing each year in Sweden. Fortunately, only about 1% of these cases are actual disappearances. The number of reported missing persons has risen dramatically during the last decade, possibly due to technical progress: such as not responding to emails or cellular phones for several days. The police investigation of a disappearance involves finding out the circumstances of the disappearance and gathering all possible information about the missing person, including the medical and dental records. If the missing person has not been found within 60 days the case becomes national and the files are sent to the police headquarters in Stockholm, where the information is put into the National Police Repository of Missing Persons and Unidentified Bodies, which is kept by the Division of the International Police Cooperation. Copies of the files with original dental records containing any/all radiographs and photographs are forwarded to the forensic odontologist for registration in the Dental Repository of Missing Persons and Unidentified Bodies.

The Disaster Victim Identification (DVI) System International, developed by Plass Data Software A/S and adopted by Interpol,[1] is used for this purpose if the cases contain any dental information. In this database the only pages that are filled out are A0, A1 and A2 as well as F1 and F2 and any photographs and radiographs are imported if they are digital or scanned and imported into the database if they are analogue. As the dental information is searchable in the DVI System with quite advanced sophistication it is extremely useful whenever a totally unknown body is found. In 2010 the latest DVI System International version (DVI3) was implemented and is being put into use by the Swedish police. In the future updates with further versions of the DVI System will be implemented continuously.

Until now almost 500 cases have been put into the new DVI System International in Sweden. Of these 64 are PM cases. Many of the PM cases have been found by other Nordic countries, most often in the waters of the Baltic Sea or Kattegat/Skagerrak (Fig.1). Also, some of the unknown deceased are apparently of foreign origin, mainly from Eastern Europe as indicated by the dental treatment.

Fig: 1. *The map of Scandinavia and the Baltic Sea.*

THE INTERPOL BLACK AND YELLOW NOTICES

When a deceased individual defies all identification effort it is time to request international assistance from Interpol. A Black Notice is then issued to the Interpol liaison offices of all member countries. The Interpol's Black Notices are routinely sent out to all 188 member countries. A Black Notice has a description of the deceased (and circumstances of the discovery) that has not been identified in the country where the remains have been found. Its purpose is to alert the police in other countries to make searches in the repositories of missing people in the hope of finding a matching identity and return the deceased to the family for appropriate disposal. The Black Notice should

also contain all necessary information that could be used for comparison with the missing persons in the national repositories of missing persons. It should include information on the dental status, with photographs and/or radiographs – if available. Unfortunately, the quality of the information varies immensely, especially concerning the dental status of the victim. At best there is a DVI F2 page written in English but photographs and/or radiographs of the dentition are a rarity. Very often it is apparent that the dental entries have not been made by a forensic dentist but a pathologist or a police officer and frequently include comments such as “full set of teeth” or “teeth in poor condition.”

The Yellow Notices are issued through Interpol for missing persons and facilitate comparisons with unknown bodies in other countries than the one of the missing person’s origin. Usually, the description of the missing person’s dentition (if present) is of acceptable quality. However, the radiographs are seldom attached; this might change in the future as digital radiographs are becoming more frequently used. Sometimes the Interpol office of the issuing country will issue a notification (sent locally to neighbouring countries only) instead of contacting the Interpol headquarters (in Lyon, France) when it seems unnecessary to bother all member countries. This is often the case (when someone has gone missing or a body has been found) in the closed off waters of the Baltic Sea.

THE DENTAL RECORD LEGISLATION

Since Dr. Oscar Amoëdo wrote the first scientific dissertation on the use of dental comparison for human identification in 1898,[2] forensic odontology has become an accepted tool in the science of identifying unknown human remains. The method has been refined continuously and with an almost universal spread of the use of dental radiography, dental identification has become extremely reliable if carried out according to appropriate scientific protocols and standards. However, the ability of the forensic odontologist to reach a conclusion is totally dependent on the availability and quality of the AM comparison material (the dental records), which in their turn are dependent on the national legislation.

Sweden is one of the most privileged countries in this respect: the medical and dental record legislation states clearly and in great detail how records should be written, kept and preserved,[3] what should be included and how to guard the integrity of computer generated records. Some of that legislation applies especially to the needs of forensic odontology. The most important laws state that records must be preserved for at least ten years after the patient’s last visit and that records must be released to the forensic pathologist or odontologist if deemed necessary to the forensic investigation. The dental profession seems to be particularly conscious of these requirements, which became apparent when 543 Swedish citizens were among the victims of the Asian Tsunami on 24th December, 2004. Of the 430 Swedish victims who were old enough to have dental records, only two records were no longer in storage. They had been destroyed because these two victims had not visited their dentists for the past 18 and 20 years respectively.

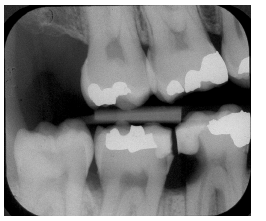
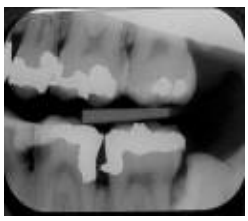
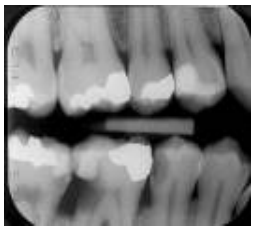
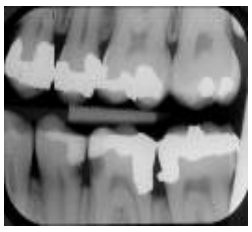
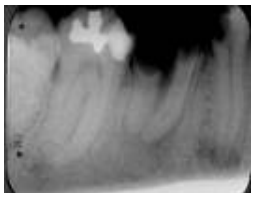
The following cases present different problems and mistakes that delayed the establishment of identity. However, these individuals were identified eventually and could be buried with their own names. Solving these cases increased knowledge and experience of all those involved in the investigation.

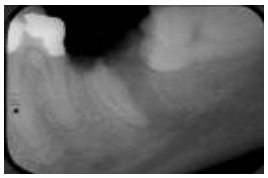
CASE 1
The man who lost three teeth.

On May 12, 2007, several pieces of a skeleton to include a cranium and mandible were found scattered in the woods outside the city of Örebro, 200 kilometres west of Stockholm. The remains with several items of clothing were brought to the forensic medicine department in Linköping, where the forensic pathologist (also a dentist) carried out an autopsy as well as the dental investigation that included photographs of the jaws and radiographs of the teeth. As the police had no idea of the identity, the results of the dental investigation were forwarded to Stockholm to be compared to the missing persons by the forensic odontologist.

However, despite several attempts, searching of the DVI System failed to yield any possible matches. To make sure that an identity would not be missed due to some mistake in the registration of the AM status in the past, a manual comparison routine was started, meaning that the unknown individual would be compared to almost every case in the repository of the missing persons. This is time consuming, undertaken only if everything else has failed. Fortunately, within a few days the police technicians investigating the artefacts (that had been found together with the skeleton) concluded that the man’s shoes were

manufactured in 2000. This information helped restrict the search range. Starting with the missing persons from 2000, the manual comparisons ended with one of the last cases from 2003. Despite several discrepancies there were enough similarities to prompt closer scrutiny. As these cases were filed in the DVI1 version there were no radiographs linked to the system. However, when the actual files were pulled from the archive and the AM and PM radiographs were compared the identity could be established with certainty.



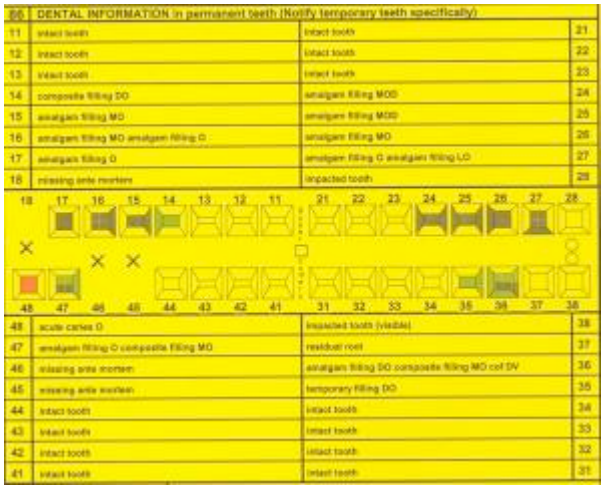
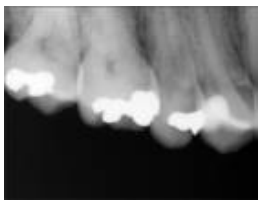
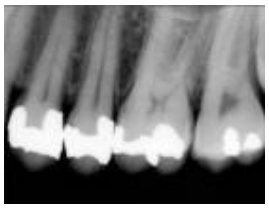
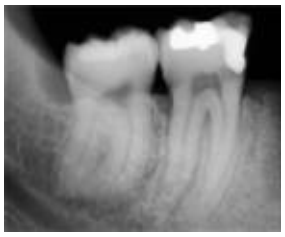
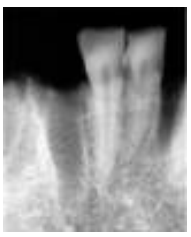
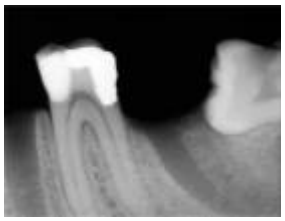


Fig. 2: CASE 1. *The radiographs and AM F2 form.*



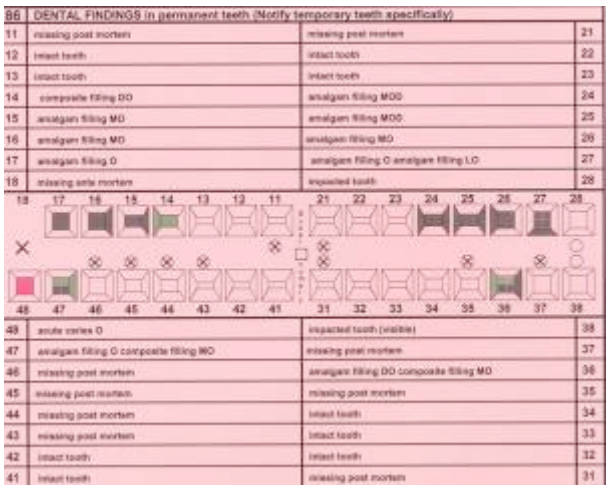


Fig. 3: CASE 1. *The radiographs and photographs and the PM F2 form of the unknown remains.*

One of the main reasons for the failure to match the cases by search or data mining was because two of the victim's teeth (45 and 46) had been extracted 16 days before he had gone missing. These teeth had been designated as missing in the AM file (Fig 2).

Further confusion was caused because tooth 37 had been resected and the distal root had been extracted three months prior to the disappearance. However, as the skeletonized remains presented empty sockets it was concluded that teeth 45 and 46 were missing PM. Additionally, the mesial root of 37 was missing PM, whereas the socket left by the distal root had healed, leading to the mistaken conclusion that the whole 37 had been lost PM (Fig. 3). The coding of the recently extracted teeth as missing AM although it might be expected that the remains, if found, would present empty sockets that were recorded as corresponding teeth missing PM was an important lesson for the author. The search engine of the DVI program had naturally excluded the correct missing individual since teeth missing AM are absolute exclusion criteria against teeth that have apparently been present at the time of death and gone missing PM. It would be more prudent to register these teeth as extracted, or write “recently extracted” as free text in the lines of the corresponding teeth.

CASE 2

What happened to the woman on the other side of the Baltic Sea?

A notification instead of a Yellow Notice was issued by the Interpol Wiesbaden on August 1, 2005. The evidence indicated that an elderly German woman had committed suicide on January 16, 2005, by jumping into the Baltic Sea off the German coast near Heringsdorf, which lies next to the German-Polish border (Fig. 1). She had left a suicide note explaining that she suffered from a disfiguring tumour disease and had decided to end her life. The nnotification contained an incompletely filled F2 form as well as a description of other characteristics. It was specifically pointed out that she was missing five front teeth in the lower jaw, which were replaced by a partial denture (Fig. 4).

On January 23, 2005, a female body was found on the beach of the city of Mrzeżyno, Poland, almost 100 km from the German border (Fig. 1). It was concluded that she had been in the water only a few days. The PM investigation showed that she had drowned. However, the Polish authorities were unable to match her to any of the missing women in Poland and the body remained unidentified. In the end a notification instead of a Black Notice was issued by Interpol Warsaw, directed to the countries at the southern end of the Baltic Sea. Among physical characteristics described, one was particularly important: a scar and evidence of tumour on the left side of her face. There was also a mention of a denture in the upper jaw (which turned out to be incorrect). An addition to that notification was created a few weeks later as demanded by the Nordic countries, including a rudimentarily filled out F2 form. The most significant information contained in the F2 form showed that six teeth in the front of the lower jaw were missing (Fig. 5). Despite several discrepancies between the German and the Polish F2 forms, the missing lower teeth as well as the evidence of facial tumour, triggered the notion that the missing German woman and the unidentified body in Poland could be the same person. A response suggesting that probability was issued through Interpol Stockholm to the German and Polish counterparts. Interpol Wiesbaden responded a few weeks later corroborating the results.

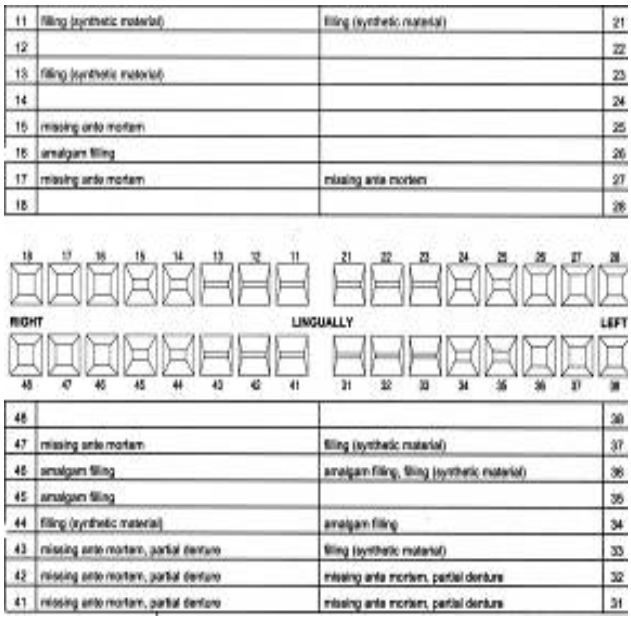


Fig. 4: CASE 2. *AM F2 form issued by Interpol Wiesbaden, Germany.*

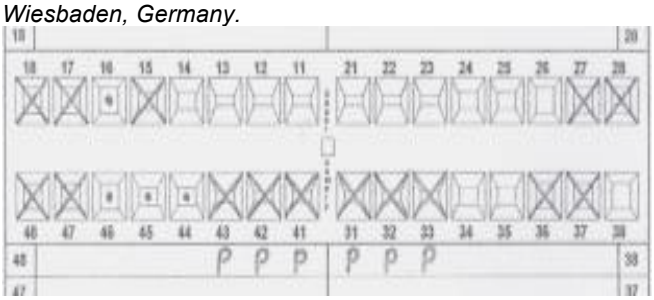


Fig. 5: CASE 2. *PM F2 issued by the Interpol Warsaw, Poland.*

As the timeline (Table 1) shows it took more than a year to establish the identity in this case despite only a few days having passed between the disappearance of the German

woman and the discovery of the deceased female on the other side of the German/Polish border. The notifications were issued many months after the actual events had taken place and the contents of the F2 forms were of poor quality (and inaccurate) in all likelihood because they had not been handled by a dentist. The comparison and conclusion of probable identity was made on the other side of the Baltic Sea almost by pure chance.

Table 1: CASE 2. *Timeline of the ID process.*

January 16, 2005	Reported missing in Germany
August 1, 2005	Notification issued (instead of Yellow Notice)
January 23, 2005	Body found in Poland
December 5, 2005	Initial notification issued (instead of Black Notice)
February 8, 2006	Additional notification with F2 issued
February 13, 2006	Comparison report issued, Stockholm: probable identity
March the 1, 2006	Confirmation of identification

CASE 3
Which way from Finland?

On the 13th of June, 2009, a badly decomposed body was found in the water near the northern Swedish city of Härnösand. It had apparently been floating in the cold waters of the northern Baltic Sea for several months: possibly it had spent the winter frozen in the ice of the Gulf of Bothnia. The body was transported to the forensic medicine department in Umeå for autopsy and identification. A full dental investigation was conducted, including radiographs and photographs of the jaws (Fig. 6). None of the proposed local cases of missing persons from the recent years matched the remains. The whole PM file of this unsolved case was forwarded to Stockholm for comparison to the entire content of the repository of the missing persons. Despite several thorough searches no match was found. Based on the detailed investigation it was concluded that the deceased person was male, somewhat taller than 180 cm and aged between 20 and 40. The details and quality of the dental treatment indicated that he was of Nordic origin, excluding the possibility that the victim was from the Baltic States or Eastern Europe. The logical next step was to contact the authorities in Finland.

Whenever there is a need of cooperation among the police forces of the five Nordic countries Interpol is usually bypassed. Information about the unresolved case was relayed to Finland from Sweden. As a result of this contact, one of the numerous odontograms that arrived showed very good concordance with the dental status of the victim. A request for radiographs yielded four bitewings in the returning mail that secured the establishment of identity (Fig. 7). The deceased man was a 37-year-old Finnish citizen who had gone missing in the tiny city of Björneborg, on the opposite side of the Bothnia Bay from Härnösand (Fig. 1). He had disappeared on November 28, 2008, and his body had travelled more than 200 km during the winter, probably moved across the sea by the ice breaking up in the spring (May in this part of the world). However, it took almost four months to solve the case, possibly because neither a Yellow Notice nor notification of disappearance was issued to the countries in the vicinity of the northern part of the Baltic Sea.

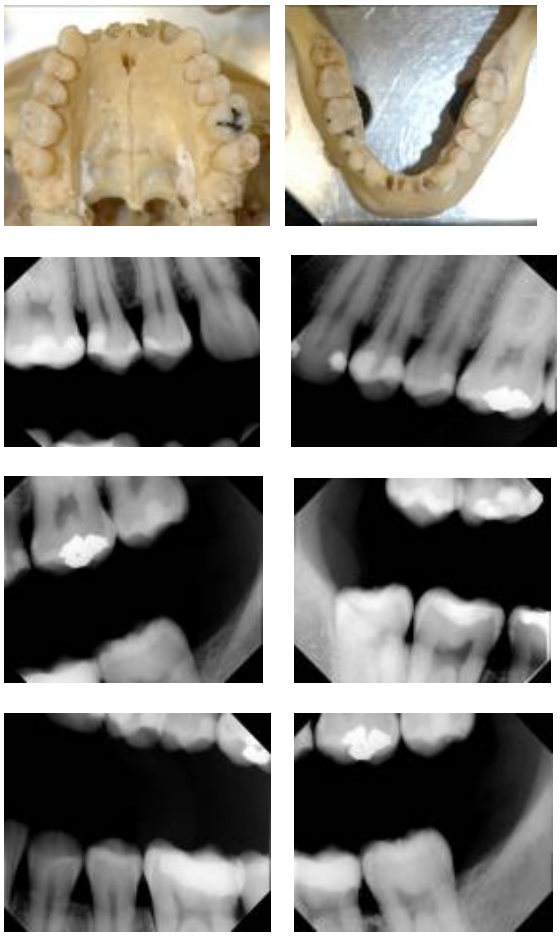


Fig. 6: CASE 3*. The PM photographs and radiographs of the unknown remains.*

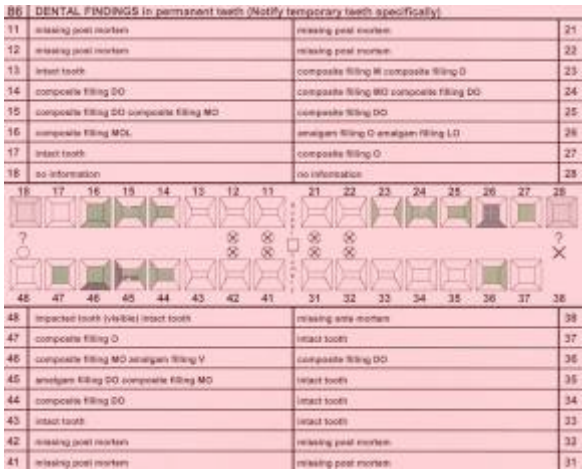


Fig. 7: CASE 3*. PM F2 form of the unknown remains.*

CONSIDERATIONS:

1, Interpretation and entry of dental information (*antemortem* and *postmortem*) to Interpol forms should always be undertaken by dental personnel.

2. Issuing the Yellow and Black Notices or notifications sooner if there is the slightest possibility that the case might have crossed any national borders.

3. Better and stricter routines for issuing the Yellow and Black Notices or notifications; compulsory use of the full sets of DVI forms including all photographs and radiographs available.

4. Additional information for any ambiguous dental situations: for example, in order to avoid exclusion by the search program describe newly extracted teeth using free text e.g. “recently extracted” instead of encoding them as MAM if the teeth that have been lost within a short period before the person had gone missing.

CONCLUSIONS

Many factors can influence the outcome of an identification investigation. Each case has its own positive and negative details that might speed up or delay identification and the return of the deceased to their families. But, as the cases described in this paper show, there are some more obvious reasons that might unnecessarily prolong the whole process. In order to avoid such delays it would be advisable to introduce and emphasise basic routines when dealing with remains without a tentative identity and missing persons. A few simple rules could save a lot of time for the professionals working on these cases and also a lot of anguish for the families of the missing individuals.

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EFFICACY OF "DIMODENT" SEX PREDICTIVE EQUATION ASSESSED IN AN INDIAN POPULATION

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ABSTRACT

Teeth are considered as a useful adjunct for sex assessment and may play an important role in constructing a post-mortem profile. The Dimodent method is based on the high degree of sex discrimination obtained with the mandibular canine and the high correlation coefficients between mandibular canine and lateral incisor mesiodistal (MD) and buccolingual (BL) dimensions. This has been evaluated in the French and Lebanese, but no study exists on its efficacy in Indians. Here, we have applied the 'Dimodent' equation on an Indian sample (100 males, 100 females; age range of 19-27yrs). Additionally, a population-specific Dimodent equation was derived using logistic regression analysis and applied to our sample. Also, the sex determination potential of MD and BL measurements of mandibular lateral incisors and canines, individually, was assessed. We found a poor sex assessment accuracy using the Dimodent equation of Fronty (34.5%) in our Indian sample, but the population-specific Dimodent equation gave a better accuracy (72%).Thus, it appears that sexual dimorphism in teeth is population-specific; consequently the Dimodent equation has to be derived individually in different populations for use in sex assessment. The mesiodistal measurement of the mandibular canine alone gave a marginally higher accuracy (72.5%); therefore, we suggest the use of mandibular canines alone rather than the Dimodent method.

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Keywords: Sex assessment; Dimodent equation; mandibular canines; mandibular lateral incisors; forensic odontology; Indians

Running Title: Dimodent equation in Indians

INTRODUCTION

Sex determination is an important aspect in post-mortem profiling and is especially useful in the identification of skeletal remains. Teeth, being resistant to a variety of post-mortem insults, are frequently used in comparative identification of human remains. The dentitions have also proved to be useful accessories in sex estimation. Additionally, since most teeth develop earlier than skeletal maturation, they can also serve as important sex determinants in young individuals.[1,2] Sex-related morphologic differences in the human skeleton have been extensively evaluated in several living and prehistoric populations. However, tooth associated odontometric differences have been less thoroughly investigated and mostly derive from the dentition of extinct populations.[3,4] Several studies have demonstrated that male teeth are generally larger than those of females in various populations.[5-11] However, the magnitudes and patterning of sexual dimorphism in permanent teeth differs from population-to-population. This could have a genetic basis and represent the cumulative effects of selection for sexual dimorphism per se.[12]

Canines, amongst all the teeth, have been shown to exhibit the maximum sexual dimorphism. They are sturdy teeth and more resistant to disease.[13] The canines are not only exposed to less plaque, calculus, abrasion from brushing, or heavy occlusal loads as compared to other teeth, but are also less severely affected by periodontal disease. Hence, they are usually the last teeth to be extracted, making them the "key teeth" for personal identification.[2,12-15]

Garn et al[12] studied sexual dimorphism by measuring the mesio-distal width of canines in different ethnic groups and concluded that the magnitude of canine tooth sexual dimorphism varied in different ethnic groups. Furthermore, the mandibular canines showed a greater degree of sexual dimorphism than the maxillary

canines.[12] Sexual dimorphism was reported to be more pronounced for molars than premolars, but posterior teeth are generally less variable than the anterior teeth. Garn et al[12] attributed this to a "field" of sexual dimorphism that includes the teeth adjacent to canines as compared with the remote teeth of the same morphologic classes. Hence, the lateral incisor tends towards greater percentage dimorphism than does the central incisor, and the first premolar shows greater percentage dimorphism than the second premolar. Thus across genera, it appears that teeth adjacent to canines share with it a tendency for greater dimorphism; the greater the canine dimorphism, the greater the sexual dimorphism of adjacent teeth.[12] The Dimodent method is thus based on the high degree of sex discrimination obtained with the mandibular canine and the high correlation coefficients between mandibular canine and lateral incisor widths. It uses logistic regression analysis based on the mesiodistal and buccolingual width of these two teeth (the mandibular canine and lateral incisor). This was first applied on a French population by Fronty et al[17] and later by Ayoub et al[18] in the Lebanese population and has given moderate to good accuracy (76.7% and 90.6%, respectively). However, the equation has not been tested on Indians. Since, it is known that sexual dimorphism varies among populations,[3,5–7,9,12,16] we additionally ventured to develop a population-specific Dimodent formula for our sample. The present study sought to test the efficacy of the Dimodent equation of Fronty as well the population-specific equation in an Indian sample.

MATERIALS AND METHODS

The present sample included dentitions from 100 males and 100 females from India belonging to the age range of 19–27 years. The sample was limited to young adults to ensure that the teeth were intact in order to obtain optimal odontometric information. Subjects were from different states of India and belonged to a mixture of ethnic groups, religions and castes so as to evaluate the sex differences in Indians in general. After obtaining verbal informed consent, maxillary and mandibular alginate impressions were made and casts were poured using dental stone.

Measurements: Mesiodistal (MD) and buccolingual (BL) dimensions of mandibular canines and mandibular lateral incisors were measured on the casts using a digital calliper with calibration of 0.01mm (Mituyoto, Japan). The MD dimension was defined as the greatest distance between the contact points on the proximal surfaces of the tooth crown. In case of tooth rotation or malposition, the measurements were obtained between points where it was considered that contact with adjacent teeth would have occurred. The BL dimension was defined as the greatest distance between the labial surface and the lingual surface of the tooth crown, measured with the calliper beaks held at right angles to the MD dimension. Measurements were repeated by a second observer on 20 randomly selected casts to test for possible inter-observer variation. Since the dimensions on right and left sides of the same dental arch are usually symmetrical,[6] the right side measurements only were taken into consideration for the present study.

Application of the Dimodent equation of Fronty: The measurements undertaken were subjected to the Dimodent sex prediction equation of Fronty et al[17] which is formulated as follows:

$P = 1/(1+e^{-y})$, where P stands for the probability of the teeth belonging to a male or female.

y is calculated as follows:

$y = 24.2 + (1.54 \cdot \text{LI-MD}) + (1.92 \cdot \text{LI-BL}) – (2.84 \cdot \text{C-MD}) – (3.38 \cdot \text{C-BL})$ (A)

LI-MD — mesiodistal dimension of the mandibular lateral incisor

LI-BL — buccolingual dimension of the mandibular lateral incisor

C-MD — mesiodistal dimension of the mandibular canine

C-BL — buccolingual dimension of the mandibular canine

According to the obtained P values, three options are possible:

1. If P tends towards 100% (i.e., P > 50%), the dentition would belong to a male
2. If P tends towards 0% (i.e., P < 50%), the dentition would be that of a female;
3. If P = 50%, discrimination is null, and sex cannot be assessed.

Deriving a population-specific Dimodent equation for Indians: The measurements were

entered into an MS Excel spreadsheet (Microsoft Corp., Redmond, Washington, USA) and logistic regression analysis (LRA) was performed for MD and BL measurements of mandibular lateral incisors and canines, as well as for the absolute measurements individually. Sex identification was determined by the following:

$P = 1/(1+e^{-y})$

Y is calculated as follows: $Y = -19.112 + (0.069 \cdot LI\text{-}MD) - (0.382 \cdot LI\text{-}BL) + (2.188 \cdot C\text{-}MD) + (1.030 \cdot C\text{-}BL)$ (B).

Statistical Analyses: The possible sex variation in MD dimensions of right mandibular canines and mandibular lateral incisors was evaluated using the independent samples t-test while the inter-observer differences were tested using the paired t-test. The LRA were performed on SPSS 16.0 software package (SPSS Inc., Chicago, Illinois, USA). The arithmetic calculations for calculating sexual dimorphism using the Dimodent equation, as well as the population-specific equation, were undertaken on an MS Excel spreadsheet.

RESULTS

Table 1 depicts the descriptive statistics and degree of sexual dimorphism for MD and BL measurements of mandibular canines and lateral incisors. We obtained statistically significant differences ($p < 0.05$) between males and females for MD dimensions of mandibular canines and mandibular lateral incisors. The paired t-test evaluating the potential inter-observer variation showed insignificant statistical differences for all the measurements (Table 2).

The accuracy of sex prediction from the Dimodent equation in the present study ranged from 3% for females to 66% for males. Overall the application of this equation was successful in sex prediction in 34.5% of the Indian sample (Table 3). The accuracy of the Indian equation ranged from 69% for males to 75% for females, with an overall success of 72% (Table 3). Additionally, the LRA of individual tooth measurements showed that the MD dimension of canine gave the best sex predictive accuracy (Table 4), followed by the BL dimension of canine, the BL dimension of lateral incisor and MD dimension of lateral incisor.

Table 1: *Descriptive statistics and t-values for the male and female mandibular canines and lateral incisors*

Variable	Female Mean (±SD)	Male Mean (±SD)	t value	p value
MD dimension of mandibular lateral incisor	5.8 (±0.35)	5.9 (±0.40)	3.174	0.002*
BL dimension of mandibular lateral incisor	5.6 (±0.49)	5.9 (±0.53)	5.354	0.000*
MD dimension of mandibular canine	6.5 (±0.37)	6.9 (±0.41)	6.639	0.000*
BL dimension of mandibular canine	6.6 (±0.55)	7.1 (±0.67)	5.729	0.000*

* Statistically significant at $p < 0.01$.

Table 2: *Descriptive statistics and t values for multiple observations*

Variable	Observer I Mean (±SD)	Observer II Mean (±SD)	Difference of Mean (±SD)	t val	p val
MD dimension of mandibular lateral incisor	5.85 (±0.47)	5.72 (±0.45)	0.13 (±0.67)	0.912	0.373
BL dimension of mandibular lateral incisor	5.94 (±0.50)	5.81 (± 0.49)	0.13 (±0.89)	0.675	0.508
MD dimension of mandibular canine	6.77 (±0.51)	6.73 (±0.55)	0.04 (±0.77)	0.269	0.791
BL dimension of mandibular canine	6.72 (±0.63)	6.61 (±0.64)	0.11 (±1.01)	0.484	0.634

DISCUSSION:

Sex determination is a first step in reconstructive identification in the field of forensics. In general, the sex of an unidentified person can be determined based on the anatomical characteristics of the external genitalia. However bones and teeth are the only available materials for sex determination in some instances such as markedly decayed or skeletonised remains. The study of teeth has been a subject of interest to anthropologists, biologists and forensic experts, as they are generally preserved even when the other bony structures have been destroyed. They are highly resistant to post-mortem insults, surviving a variety of destructive effects caused by trauma and incineration. Sexual dimorphism in tooth size has been explored recently with most authors concentrating on the use of BL and MD dimensions. These are easy to obtain and have demonstrated high degrees of sexual dimorphism in various studies.[8,9,12,14]

Furthermore, it is reported that tooth size is greatly influenced by genetics. Therefore, such measurements are considered to be population-specific and do not apply to the world at large.[12] Such odontometric data in sexing, especially in this part of Asia, has not been extensively studied. The present study sought to apply the Dimodent sex predictive equation given by Fronty et al[17] (A) on an Indian sample to test its efficacy. The overall accuracy of this equation in our sample was only 34.5%, with 66% of males and a mere 3% of females being correctly identified. In comparison, this equation gave a high predictive accuracy of 90.6% in the French[17] and moderate accuracy in the Lebanese (76.7%)[18] demonstrating the population-specific differences in the tooth dimensions as well as sexual dimorphism in various populations. The equation especially showed a poor accuracy in sexing females in our sample, and is similar to the findings of Acharya and Mainali,[2] who attributed it to relatively larger tooth dimensions of females. This could have resulted in poor identification of females when compared to previous studies, thus decreasing the overall sex discrimination potential of the equations.[2] This is corroborated by Prabhu and Acharya,[6] who said that the magnitude of sexual dimorphism in tooth dimensions in Indians is lower when compared to populations from other continents. This could be due to varied interactions between different genetic and environmental factors, resulting in large variations in the magnitude of sexual dimorphism across populations. Consequently, the Dimodent equation of Fronty et al[17] established on a French population gave poor sex assessment accuracy in Indians.

Therefore, we derived a Dimodent equation for our sample using LRA. This population-specific equation (B) was then evaluated on our sample, which vastly improved predictive accuracy to 72%; with 75% of males and 69% of females being correctly identified (Table 3).

Dimodent equation of Fronty et al[17]	66/100	66	3/100	3	69/200	34.5
Present sample	75/100	75	69/100	69	144/200	72%

Additionally, each measurement of both teeth was also analysed with LRA. Surprisingly, the MD measurement of canines alone gave a predictive accuracy of 72.5% with 77 % of males and 68% of females being correctly identified (Table 4). This was marginally greater to that of the population-specific Dimodent equation (B) (Table 3) and suggests that the magnitude of sex differences in the Dimodent equation comes predominantly from the canine and the use of other teeth may result in a slightly lower accuracy. Canines differ from other teeth with respect to survival and sex dichotomy. These differences are probably related to their function, which is different on an evolutionary basis from other teeth.[2,13] It is postulated that canines are not primarily masticatory in function but are usually related to threat of aggression especially in primates. In humans, this aggressive function was gradually transferred to fingers from teeth; however, until then, males largely depended on canines for their survival. Consequently, in modern humans too, it is not by chance that canines are the teeth that demonstrate the maximum sexual dimorphism.[2,8,12,18–20] While the use of canines alone makes sex estimation straightforward and expedient, the efficacy may be of moderate nature (72%). Indeed, a recent study by Acharya et al[21] has shown high sex identification accuracy (100%) in Indians using LRA of all teeth and demonstrated its potential for use as the sole indicator of sex. As an analogy, the use of all teeth in our sample may further increase sex predictive accuracy, which we intend to explore in the near future.

Table 3: *Comparison of the accuracy of sexing the present sample using Dimodent equation of Fronty and population-specific dimodent equation*

Equation	Males		Females		Total	
	N	%	N	%	N	%

Table 4: *Sex classification accuracy using logistic regression analysis of individual teeth.*

Variable	Male		Female		Total	
	N	%	N	%	N	%

MD dimension of mandibular lateral incisor	60/100	60	54/100	54	114/200	57
BL dimension of mandibular lateral incisor	59/100	59	59/100	59	118/200	59
MD dimension of mandibular canine	77/100	77	68/100	68	145/200	72.5
BL dimension of mandibular canine	63/100	63	63/100	63	126/200	63

CONCLUSION

This study supports the notion that sexual dimorphism in teeth is population-specific as the application of Dimodent equation of Fronty et al[17] gave very poor sex classification accuracy (34.5%) in Indians. The population-specific Dimodent equation, however, gave an accuracy of 72% suggesting that, if used, the Dimodent equation needs to be population-specific. Further, in our study, it was revealed that LRA using MD dimensions of mandibular canines alone gave marginally better results to that of the population-specific Dimodent equation, suggesting that canines be used independently (rather than the use of the Dimodent equation and the method therein of using mandibular lateral incisors with canines). But even this is still suboptimal (72.5%) and the use of all teeth (when present) may be better suited for sex prediction in forensic contexts with these methods.

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BOOK REVIEW

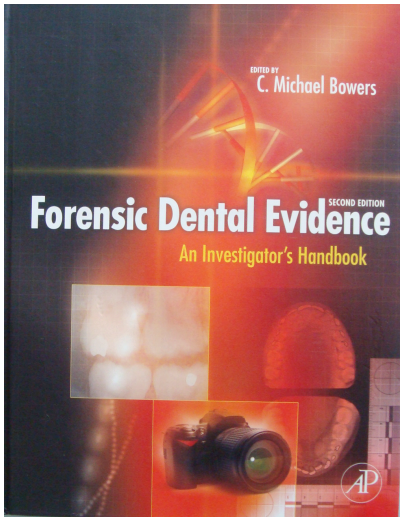
Forensic dental Evidence:
An Investigator's Handbook 2nd ed.

Edited by C. Michael Bowers
Elsevier 2011
ISBN: 978-0-12-382000-6

The latest edition of this book, comprising 16 chapters, has contributions from international authors with extensive experience in the field of forensic odontology. The purpose of this book is to give a detailed overview of modern day forensic odontology targeting those agencies investigating cases where dental evidence is discovered. Additionally, it is an extremely useful, interesting and educational compilation for anyone connected with forensic dental casework, such as coroners, medical examiners, forensic scientists, prosecutors, defence teams or those with an interest in the subject; dental/medical students, dentists and forensic odontologists (current and future) whatever their experience level.

It expands upon and updates the previous edition and draws attention to the fundamental methodologies and protocols necessary for successful casework with multi-disciplinary involvement. Chapters are colour coded for easy reference and cover topics such as historical dental investigations, identification, mass fatality incidents, bite marks, legal systems, abuse issues, dental materials in identification, DNA, photography, digital imaging and much more of interest. There is a strong emphasis on teamwork and the recognition, documentation, collection, preservation and interpretation of dental evidence. It highlights the challenges and limitations of forensic dental evidence, with a cautionary tone with regard to bite marks and mentions the work of the Innocence Project.

One chapter includes basic dental terminology, numbering and descriptions for those not familiar with dentistry. All chapters have clear sub-headings, are very well illustrated and contain fascinating historical and contemporary cases to include a Romano-Egyptian red-shroud mummy dating to the first century AD, a missing pilot discovered in 2007,



Australian bushfires 2009, bite marks, Ted Bundy and much more. It is extremely easy to read, without lengthy pages of uninterrupted text.

The electronic version of this book contains links enabling the reader to easily access all reference abstracts and updated web-based material online, encouraging further reading and research on forensic dentistry and related scientific disciplines. Very impressive!

Dental age estimation is not mentioned; perhaps there was a reason for this omission, but I feel its inclusion would have been useful. Judicial requirements, identification and persons without documentation often involve the establishment of age. There is much information and research in this area and a chapter dedicated to its use and limitations might be beneficial.

In conclusion this book comprehensively covers concepts and protocols necessary for criminal investigation involving dental evidence and will be of educational value and interest to many categories of reader. It is practical, up to date, informative, well-illustrated and appropriately cautionary.

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